IND ——
CROSSREFERENCE TO IND ——

Review Completed: December 1, 1998

Sponsor: Sensus Drug Development Corp.; Austin, TX 78701

Date Submitted: November 20, 1998 **Date Received:** November 23, 1998

PHARMACOLOGY REVIEW OF IND SUBMISSION
IND ——— (November 20, 1998)

DRUG: Trovert™; B2036-PEG; rhGH antagonist (PEGylated)

STRUCTURAL FORMULA: B2036-PEG is a 191 amino acid polypeptide with two disulfide bonds. As an analogue of hGH, it contains nine substitutions in the first and third α-helices. There are 4-5 PEG moieties (~5000 Da each) covalently bound to amino groups on the surface of the molecules. This increases its mass to 42-46,000Da.

FORMULATION: Per vial: 10 mg B2036 protein. Mannitol (36 mg); glycine (1.36 mg) and —— sodium phosphate, pH 7.4. To be reconstituted in water up to 10 mg/ml (for clinical testing).

CATEGORY: GH antagonist, PEGylated

INDICATION: Treatment of acromegaly (third line).

RELATED IND/NDA: IND —— NDA 19-667 (Sandostatin).

MEMOS: Pre-IND meeting held with sponsor January 24, 1996. Alex Jordan and Ron Steigerwalt were pharmacologists present. Key toxicology items discussed: Dr. Jordan indicated that the monkey studies would probably be sufficient and also noted that carcinogenicity studies would eventually be required. Teratology should be performed in a species in which the drug works. There was a meeting with the sponsor regarding preclinical requirements for both acromegaly and —— indications. The CDER reproductive toxicology committee was consulted as to the appropriate design of studies. Joe DeGeorge was consulted regarding carcinogenicity studies. For Acromegaly, the Reproductive Toxicology committee recommended a modified Segment I/Segment II approach. The results of these consults were incorporated into a report of the minutes of the May 22, 1998 meeting (see copy of a summary of conclusions from the minutes from May 22, 1998 meeting with the sponsor. These comments were communicated to the sponsor in a fax on September 17, 1998.

CLINICAL STATUS: Phase 2: Protocol # SEN-3611: A Six Week, Double Blind, Placebo Controlled, Multiple Dose, Phase IIb study of B2036-PEG in the Treatment of Acromegaly. 45 patients (15/group) will receive weekly subcutaneous administrations for 6 weeks. Dose groups listed in protocol are 0, 30 and 80 mg of B2036-PEG. (Elsewhere in the submission, the dose selection is described as depending upon previous human experience). This was the initial plan, but the sponsor has since switched to daily dosing of 20 mg, with the possibility of increasing the dose further depending upon the patient response.

PREVIOUS HUMAN EXPERIENCE: Phase I: Placebo controlled, double blind, single rising dose study in healthy male volunteers was "nearing completion" at time of submission. B2036-PEG was "well-tolerated at doses of up to 1.0 mg/kg and efficacious in suppressing IGF-1 concentrations" at 0.3 mg/kg (3/6 pts) and 1.0 mg/kg (6/6 pts). No serious AE's; Other AE's resolved without

sequelae. PK appeared to be linear within the 0.03 and 0.1 mg/kg groups. The PK were non-linear at effective doses of 0.3 and 1.0 mg/kg.

A single dose study in six acromegalic patients was initiated at ——— The ——— No data are available at this time.

Additional studies have assessed the daily administration of 20 mg/day in Acromegalic patients.

AE's: Preliminary data available:

0.3 mg/kg: (total 3 AE's)
1 subject unifocal ventricular extrasystoles before and after dosing. Extrasystoles remained 7 days later. Unlikely relationship with drug.

1 subject Abdominal pain and diarrhea (mild) Listed as remotely related to drug.
1 subject Headache (moderate). Listed as possibly related to drug.

0.1 mg/kg: 9 AE's: All mild: GI fullness, chest pressure, decreased appetite and headache considered remotely related to drug. Abdominal discomfort (possibly related) and nasal congestion and abnormal hunger were considered not related.

0.3 mg/kg: 3 AE's in 1 subject: rhinitis (day -3), oral herpes lesion (day -1). Mild itching at injection site lasting 12 h probably related to drug.

1.0mg/kg: 14 AE's: moderate epistaxis after minor trauma (remotely related) and headache (not related) 2 headaches were considered possibly related and parathesias of the tongue (2). Dry throat (2) and abdominal pain (1) were considered remotely related.

REVIEW OF ——— STUDY NUMBER 0301FS35.001:
AMES/SALMONELLA-E.COLI REVERSE MUTATION ASSAY ON B2036-PEG

Note: Study performed by ——— Study completed: 7/21/98. Signed GLP and QA statements were provided.

Purpose: *In Vitro* assessment of mutagenic potential of B2036-PEG using a bacterial reversion assay ("Ames Test").

Experimental Design: Preliminary toxicity screen utilized both pre-incubation and plate incorporation methods. This screen utilized TA1537, TA100, and WP2*uvrA* at doses up to 5000 µg/plate with sterile water for vehicle. No metabolic activation was used in the preliminary screen. Test agent was not toxic and freely soluble at all concentrations.

Definitive study utilized both plate incorporation and liquid pre-incubation conditions. The six tester strains were tested at doses of 16.7, 50.0, 167, 500, 1670 and 5000 µg/plate with and without metabolic activation in both assay types. Preincubation was for 30 min at 30°C. Plates from both methods were incubated at 37 °C for 2 days. Colonies were counted automatically with an ——— electronic colony counter.

Salmonella Strains: TA1535, TA1537, TA98, TA100, TA102
E. coli strain: WP2*uvrA*

Metabolic Activation System: Standard S9 from Aroclor-induced male SD rat liver homogenate. (6%v/v).

Test article: B2036-PEG (lot#372753A): 16.7, 50.0, 167, 500, 1670 and 5000 µg/plate ± metabolic activation.

Vehicle control: Sterile water.

Positive control without S9:
TA1535 and TA100: sodium azide (10.0 µg/plate).
TA1537: 9-aminoacridine (150 µg/plate).
TA98: 2-nitrofluorene (5 µg/plate).
TA102: mitomycin C (2.5 µg/plate).
WP2*uvrA*: ENNG (2.0 µg/plate).

Positive control with S9:
TA102: 2-Aminofluorene (30 µg/plate).
TA1535, TA1537, TA100, TA98: 2-anthramine (2.5 µg/plate).
WP2*uvrA*: 2-anthramine (80 µg/plate).

Criteria for positive result: Statistically significant, dose-dependent increase in the number of revertants with at least one dose level inducing a revertant frequency that is 2-fold the concurrent negative control value.

Results: Results in both preliminary and definitive tests comprise a clear negative result for this test. There were no increases in colony counts with the test agent for any strain under any test condition. Positive controls produced expected results indicating a valid test.

Evaluation: This is a valid test performed at the maximum recommended doses for soluble, non-toxic comp ictivity under the conditions c

Summary Data - Liquid Pre-incubation Assay

CONTROLS

AVERAGE REVERTANTS/PLATE

SOLVENT CONTROLS	S9	TA1535	TA1537	TA98	TA100	TA102	UVR A
PLACEBO (250 UL)	(-)	8 (1)	9 (1)	26 (4)	288 (24)	299 (54)	6 (2)
PLACEBO (250 UL)	(+)	7 (2)	14 (2)	34 (8)	258 (70)	384 (8)	9 (5)
POSITIVE CONTROLS (UG/PL)							
SODIUM AZIDE 10.0	(-)	1842*(102)	-	-	919*(57)	-	-
9-AMINOACRIDINE 150	(-)	-	1275*(187)	-	-	-	-
2-NITROFLUORENE 5.00	(-)	-	-	639*(78)	-	-	-
MITOMYCIN C 2.50	(-)	-	-	-	-	1401*(24)	-
ENNG 2.00	(-)	-	-	-	-	-	823*(126)
2-ANTHRAMINE 2.50	(+)	197*(54)	288*(37)	1523*(410)	1491*(163)	-	-
2-AMINOFLUORENE 30.0	(+)	-	-	-	-	1244*(117)	-
2-ANTHRAMINE 80.0	(+)	-	-	-	-	-	619*(75)

TEST ARTICLE: B2036-PEG

AVERAGE REVERTANTS/PLATE

DOSE LEVEL (UL/PL)	S9	TA1535	TA1537	TA98	TA100	TA102	UVR A
16.7	(-)	6 (3)	12 (6)	25 (4)	261 (26)	305 (90)	5 (1)
50.0	(-)	8 (1)	7 (7)	24 (3)	251 (15)	262 (26)	8 (6)
167	(-)	9 (3)	9 (2)	32 (6)	228 (25)	258 (34)	6 (1)
500	(-)	10 (4)	10 (3)	26 (6)	259 (31)	242 (38)	3 (0)
1670	(-)	13 (1)	6 (2)	19 (6)	299 (59)	296 (92)	7 (0)
5000	(-)	5 (2)	7 (2)	22 (3)	248 (31)	247 (66)	6 (1)
16.7	(+)	6 (1)	9 (3)	38 (2)	336 (41)	361 (78)	9 (4)
50.0	(+)	11 (4)	13 (3)	27 (3)	292 (12)	401 (25)	7 (5)
167	(+)	12 (5)	9 (7)	42 (4)	318 (19)	327 (53)	6 (2)
500	(+)	10 (3)	11 (2)	39 (6)	301 (38)	328 (44)	4 (3)
1670	(+)	9 (3)	12 (3)	37 (6)	300 (23)	406 (119)	5 (1)
5000	(+)	6 (2)	14 (3)	29 (9)	268 (47)	371 (38)	5 (1)

*Positive Response: 2X Solvent (TA1535, TA1537, TA98, TA100, TA102, UVR A).
Data Reported as: Mean (Standard Deviation)
Apparently normal growth all strains/doses ±S9.
Test article freely soluble at all doses.

Id W. Steigerwalt
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REVIEW AND EVALUATION OF PHARMACOLOGY/TOXICOLOGY DATA:

KEY WORDS: Pharmacokinetics, Acute Toxicology, Multiple dose Toxicology

Reviewer Name: Ronald W. Steigerwalt, Ph.D. Pharmacology Team Leader
Division Name: Division of Metabolic and Endocrine Drug Products (DMEDP)
HFD#510
Review Completion Date: February 24, 1999
Review number: 4

IND/NDA NUMBER: IND ——
Serial number/date/type of submission: SN#034, January 26, 1999 IT
Information to sponsor: Yes (X) No ( )
Sponsor (or agent): Sensus Drug Development Corp.; Austin, TX 78701

DRUG
Code Name: B2036-PEG
Generic Name: rhGH antagonist (PEGylated)
Trade Name: Trovert™
Molecular Formula/ Molecular Weight: B2036-PEG is a 191 amino acid polypeptide with two disulfide bonds. As an analogue of hGH, it contains nine substitutions in the first and third α-helices. There are 4-5 PEG moieties (~5000 Da each) covalently bound to amino groups on the surface of the molecules. This increases its mass to 42-46,000Da.

Relevant INDs/NDAs/DMFs: IND ——

Drug Class: Modified recombinant human growth hormone.

Indication: Acromegaly (third line treatment)

Clinical formulation: Per vial:
10 mg B2036 protein
36 mg Mannitol
1.36 mg glycine
—— sodium phosphate, pH 7.4.
To be reconstituted in water up to 10 mg/ml (for clinical testing).

Route of administration: SC daily

Proposed clinical protocol or Use: No new protocols proposed in this submission. Initial studies attempted weekly administration of 80 mg. This has been increased to daily administration of 20 mg/day (total 140 mg/week). Individual unresponsive patients have received higher doses on a daily basis.

Previous clinical experience: Phase I: Placebo controlled, double blind, single rising dose study in healthy male volunteers was "nearing completion" at time of submission. B2036-PEG was "well-tolerated at doses of up to 1.0 mg/kg and efficacious in suppressing IGF-1 concentrations" at 0.3 mg/kg (3/6 pts) and 1.0 mg/kg (6/6 pts). No serious AE's; Other AE's resolved without sequelae. PK appeared to be linear within the 0.03 and 0.1 mg/kg groups. The PK were non-linear at effective doses of 0.3 and 1.0 mg/kg.

A single dose study in six acromegalic patients was initiated at ——— The ——— No data are available at this time.

Additional studies have assessed the daily administration of 20 mg/day in Acromegalic patients.

AE's: Preliminary data available:

0.4 mg/kg: (total 3 AE's)
1 subject unifocal ventricular extrasystoles before and after dosing. Extrasystoles remained 7 days later. Unlikely relationship with drug.

1 subject Abdominal pain and diarrhea (mild) Listed as remotely related to drug.
1 subject Headache (moderate). Listed as possibly related to drug.

0.1 mg/kg: 9 AE's: All mild: GI fullness, chest pressure, decreased appetite and headache considered remotely related to drug. Abdominal discomfort (possibly related) and nasal congestion and abnormal hunger were considered not related.

0.3 mg/kg: 3 AE's in 1 subject: rhinitis (day -3), oral herpes lesion (day -1). Mild itching at injection site lasting 12 h probably related to drug.

1.0mg/kg: 14 AE's: moderate epistaxis after minor trauma (remotely related) and headache (not related) 2 headaches were considered possibly related and parathesias of the tongue (2). Dry throat (2) and abdominal pain (1) were considered remotely related.

Phase 2: Protocol # SEN-3611: A Six Week, Double Blind, Placebo Controlled, Multiple Dose, Phase IIb study of B2036-PEG in the Treatment of Acromegaly. 45 patients (15/group) will receive weekly subcutaneous administrations for 6 weeks. Dose groups listed in protocol are 0, 30 and 80 mg of B2036-PEG. (Elsewhere in the submission, the dose selection is described as depending upon previous human experience). This was the initial plan, but the sponsor has since switched to daily dosing of 20 mg, with the possibility of increasing the dose further depending upon the patient response.

Disclaimer – use of sponsor's material: No sponsor material is reproduced in this report.

INTRODUCTION AND DRUG HISTORY:
Pre-IND meeting held with sponsor January 24, 1996. Alex Jordan and Ron Steigerwalt were pharmacologists present. Key toxicology items discussed: Dr. Jordan indicated that the monkey studies would probably be sufficient and also noted that carcinogenicity studies would eventually be required. Teratology should be performed in a species in which the drug works. There was a meeting with the sponsor regarding preclinical requirements for both acromegaly ——— indications. The CDER reproductive toxicology committee was consulted as to the appropriate design of studies. Joe DeGeorge was consulted regarding carcinogenicity studies. For Acromegaly, the Reproductive Toxicology committee recommended a modified Segment I/Segment II approach. The results of these consults were incorporated into a report of the minutes of the May 22, 1998 meeting (see copy of a summary of conclusions from the minutes from May 22, 1998 meeting with the sponsor. These comments were communicated to the sponsor in a fax on September 17, 1998.

The present submission provides some additional studies with validation of the RIA for B2036-PEG. Final study reports are provided for some of the studies that were initially reviewed under

the initial submission. Additional PK data from the toxicology studies. These reviews are reproduced in this review with the PK data appended.

Studies reviewed within this submission: Most studies were submitted in the initial IND submission and have been previously reviewed as draft studies. The final studies include PK data. The initial reviews are reproduced in this review with the addition of PK data. There were no differences in the toxicology sections of the reports in the finalized versions compared to the drafts.

Studies not reviewed within this submission: SEN-002: The quantitative determination of B2036-PEG in mouse serum by RIA SEN-003
(samples evaluated from Studies SEN-104 Acute IV study in mice and SEN-110 14-Day IV Toxicity Study in Mice.). The results of this study are reported in the individual toxicity studies as a PK component of the study.

TOXICOLOGY:

General Comments: The following toxicity studies were reviewed under the original IND submission. They are reproduced in the new pharmacology review format with the addition of Toxicokinetic data where applicable.

Study Title: **Acute Intravenous Toxicity Study In Mice**
Study No: SEN 103
Amendment #034, Vol. 2 #, and page #: IND page # not provided.
Conducting laboratory and location:
Date of study initiation: February 9, 1996
GLP compliance: Yes
QA- Report Yes (X) No ()

METHODS:
Dosing: Vehicle (Nutropin Buffer), or 0.3, 1.0, 3.0, and 10 mg/kg B2306-PEG via the tail vein. The remaining mice were observed at 1, 4 and 24 h post-dose, then daily thereafter for a total of 14 days. On day 15, gross necropsy was performed. Statistics: ANOVA by Dunnett's procedure.
species/strain: CD-1 mice
#/sex/group or time point: 5/sex/group
age: 5-6 weeks rather than the generally accepted 6-12 weeks
weight: 19-28 g
satellite groups used for toxicokinetics or recovery: Three additional groups (2/sex/group) received 0.3 or 10 mg/kg B2036PEG or no treatment. The latter groups were anesthetized and bled approximately 1 hour post-dose.
dosage groups in administered units: Vehicle (Nutropin Buffer), or 0.3, 1.0, 3.0, and 10 mg/kg B2306-PEG via the tail vein.
route, form, volume, and infusion rate: 10 ml/kg IV
Drug, lot#, radiolabel, and % purity: lot number not provided.

Formulation/vehicle: Nutropin buffer

OBSERVATIONS AND TIMES:
Clinical signs: Mortality observed twice daily. Clinical signs observed 1, 4 and 24 h after dose, then daily.
Body weights: Day of randomization, day 1 prior to dose then days 8 and 15.
Hematology: In satellite groups by cardiocentesis on day 1, in groups 1-5 on day 15. HGB, HCT, RBC, WBC, PLT, MCV, MCH, MCHC.
Clinical chemistry: Creat, Tot Prot, alb, glob, A/G ratio, glu, ALT, AST, tot chol, CK, BUN, TBILI, ALP, Na, Phos, Ca, Cl, Chol, K
Gross pathology: terminal.
Toxicokinetics: From day 1 and 15

RESULTS:

Clinical signs: No clinical signs or mortality.

Body weights: Sponsor reports an increase in mean body weight for all test animals. Data indicate that this apparently refers to the normal growth of animals (including controls) during the experiment. There were no statistically significant changes in body weights in any groups that were related to treatment.

Hematology: Statistically significant decrease in total and differential leukocyte counts (WBC) in males at the low and high dose groups compared to vehicle control (WBC: 14.22, 8.65*, 9.58, 11.64 and 9.04* $10^3/mm^3$ for control, L, M1, M2 H; $p<0.05$). Significant decrease in lymphocyte counts were also observed in mid and high dose males (11.09, 6.87*, 7.59, 8.91, 6.3* $10^3/mm^3$ for control, L, M1, M2 H; $p<0.05$). Statistically significant decrease in basophils and leukocytes in M1 dose males (not dose dependent or likely to be biologically significant). There was a large variability in neutrophil count ($10^3/mm^3$) in high dose males due primarily to a single high dose male that had a reading of 5.74 $10^3/mm^3$ while the other animals were in the 0.85-2.35 range. No significant differences were noted in any female groups. For both sexes, there was a slight, non-statistically significant decrease in monocytes with increasing dose. This is not likely to be biologically significant, but was notable in that it occurred in both sexes. Polychromasia was observed in all groups, including vehicle (perhaps a characteristic of the younger mice?).

Clinical chemistry: Statistically significant increase in globulin in M2 dose males (not dose related).

Gross pathology: No treatment-related findings.

Toxicokinetics: B2036-PEG was systemically available. Serum concentrations were greater after administration of 10 mg/kg than 0.3 mg/kg (~40X). Mean serum concentrations were 4446 and 168, 449 ng/ml for the 0.3 and 10 mg/kg doses, respectively.

KEY STUDY FINDINGS:
1. Statistically significant decrease in total and differential leukocyte counts (WBC) in males at the low and high dose groups compared to vehicle control (WBC: 14.22, 8.65*, 9.58, 11.64 and 9.04* 103/mm3 for control, L, M1, M2 H; $p<0.05$). Significant decrease in lymphocyte counts were also observed in mid and high dose males (11.09, 6.87*, 7.59, 8.91, 6.3* 103/mm3 for control, L, M1, M2 H; $p<0.05$).
2. There were no clear dose-related toxicities observed in this acute toxicity study in mice. Doses up to 10 mg/kg were well tolerated acutely. Based on surface area comparisons to the

proposed daily dosing of 20 mg/day in humans, this represents a multiple of human exposure of ~ 3X.

Study Title: **Acute Intravenous Toxicity Study In CD-1 Mice (basically a repeat of above study)**
Study No: SEN-104;
Amendment #034, Vol #2, and page #: IND page numbers not provided
Conducting laboratory and location: :
Date of study initiation: August 7, 1996
GLP compliance: Yes
QA- Report Yes (X) No ()

METHODS:
Dosing: Single IV dose of vehicle (Nutropin Buffer), or 0.3, 1.0, 3.0, and 10 mg/kg B2306-PEG via the tail vein. Dose volume was 10 ml/kg. Three additional groups (2/sex/group) received 0.3 or 10 mg/kg B2036PEG or no treatment. The latter groups were anesthetized and bled approximately 1 hour post-dose.
species/strain: CD-1 mice
#/sex/group or time point: 5/sex/group
age: 5-6 weeks
weight: 19-28 g
satellite groups used for toxicokinetics or recovery: Three additional groups (2/sex/group) received 0.3 or 10 mg/kg B2036PEG or no treatment. These were anesthetized and bled by cardiocentesis ~1h post dose.
dosage groups in administered units: Vehicle (Nutropin Buffer), or 0.3, 1.0, 3.0, and 10 mg/kg B2306-PEG via the tail vein.
route, form, volume, and infusion rate: 10 ml/kg IV
Drug, lot#, radiolabel, and % purity: Not provided.
Formulation/vehicle: Nutropin buffer

OBSERVATIONS AND TIMES:
Clinical signs: Mortality observed twice daily. Clinical signs observed 1, 4 and 24 h after dose, then daily.
Body weights: Day of randomization, day 1 prior to dose then days 8 and 15.
Hematology: In satellite groups by cardiocentesis on day 1, in groups 1-5 on day 15. HGB, HCT, RBC, WBC, PLT, MCV, MCH, MCHC.
Clinical chemistry: Creat, Tot Prot, alb, glob, A/G ratio, glu, ALT, AST, tot chol, CK, BUN, TBILI, ALP, Na, Phos, Ca, Cl,Chol, K
Gross pathology: terminal.
Toxicokinetics: From day 1 and 15

RESULTS:

Clinical signs: No mortality, no clinical signs.

Body weights: Sponsor reports an increase in mean body weight for all animals. Data indicate that this apparently refers to the normal growth of animals (including controls) during the experiment. There were no statistically significant changes in body weights in any groups that were related to treatment.

Food consumption: No data.

Hematology: Statistically significant increase in MCHC in all treated female groups compared to controls (33.3, 34.2*, 34.0*, 33.9*, and 34.1%* for control, L, M1, M2, HD, respectively). Polychromasia was noted in all groups. Anisocytosis was observed in the M2 and HD females, but were within historical control ranges. There was a slight, non-significant dose-related trend to increased WBC counts in both sexes.

Clinical chemistry: Significant increase in sodium and chloride in the M1 males and increase in Cl^- in M1 and M2 males.
Na^+: 146, 147, 149*, 148, 145 (control, L, M1, M2, HD) (*p<0.01)
Cl^-: 101, 104, 107*, 106*, 103 (control, L, M1, M2, HD) (*p<0.01)
These changes were not dose-dependent.
No significant findings were determined in females.

Gross pathology: No treatment-related findings.

Toxicokinetics: B2036-PEG was systemically available. Serum concentrations were greater after administration of 10 mg/kg than 0.3 mg/kg. (~30X). Mean serum concentrations were 5599 and 187715 ng/ml for the 0.3 and 10 mg/kg doses, respectively

KEY STUDY FINDINGS:
1. There was a slight, non-significant dose-related trend to increased WBC counts in both sexes. While non-significant, this is consistent with the previous study.
2. There were no clear dose-related toxicities observed in this acute toxicity study in mice. Doses up to 10 mg/kg were well tolerated acutely. Based on surface area comparisons to the proposed daily dosing of 20 mg/day in humans, this represents a multiple of human exposure of ~ 3X.

Study Title: Acute Subcutaneous Toxicity Study In Mice
Study No: SEN-105

Amendment #034, Vol # 2, and page #: IND page numbers not provided.
Conducting laboratory and location:
Date of study initiation: February 1, 1996
GLP compliance: Yes
QA- Report Yes (X) No ()

METHODS:
Dosing: A single dose of vehicle (Nutropin Buffer), or 0.3, 1.0, 3.0, and 10 mg/kg B2306-PEG SC 5 ml/kg. The remaining mice were observed at 1, 4 and 24 h post-dose, then daily thereafter for a total of 14 days. On day 15, gross necropsy was performed.
species/strain: CD-1 mice
#/sex/group or time point: 5/sex/group
age: age 5-6 weeks
weight: 19-28 g
satellite groups used for toxicokinetics or recovery: Three additional groups (2/sex/group) received 0.3 or 10 mg/kg B2036PEG or no treatment. The latter groups were anesthetized and bled by cardiocentesis approximately 1 hour post-dose.
dosage groups in administered units: 0, 0.3, 1.0, 3.0, and 10 mg/kg B2306-PEG SC
route, form, volume, and infusion rate: 5 ml/kg SC
Drug, lot#, radiolabel, and % purity: Not provided.
Formulation/vehicle: Nutropin buffer.

OBSERVATIONS AND TIMES:
Clinical signs: Mortality observed twice daily. Clinical signs observed 1, 4 and 24 h after dose, then daily.
Body weights: Day of randomization, day 1 prior to dose then days 8 and 15.
Hematology: In satellite groups by cardiocentesis on day 1, in groups 1-5 on day 15. HGB, HCT, RBC, WBC, PLT, MCV, MCH, MCHC.
Clinical chemistry: Creat, Tot Prot, alb, glob, A/G ratio, glu, ALT, AST, tot chol, CK, BUN, TBILI, ALP, Na, Phos, Ca, Cl,Chol, K
Gross pathology: terminal.
Toxicokinetics: From day 1 and 15

RESULTS:

Clinical signs: No clinical signs, no mortality.

Body weights: Sponsor reports an increase in mean body weight for all test animals. Data indicate that this apparently refers to the normal growth of animals (including controls) during the experiment. There were no statistically significant changes in body weights in any groups that were related to treatment.

Hematology: No treatment related findings. Statistically significant increase in MCHC for M1 and M2 males (32.3, 33.0, 33.7*, 33.5*, 32.9%, $p \leq 0.01$). Also, an increased eosinophil% in M1 males (0.8±0.48, 1.2±0.28, 2.4*±1.18,1.6±1.05,1.3±0.24 $p < 0.05$). Significant decreases in neutrophils for M1 and M2 females (204, 1.42, 1.13*, 1.11*,1.51, $p<0.01$). Increase in lymphocyte % in M2 females (78.5, 81.6, 80.1, 85.8, 82.8, $p<0.01$). Polychromasia was noted in all groups.

Clinical chemistry: Statistically significant increase in Cl- for M1 males (108, 109, 112*, 110, 108, $p<0.05$). Sponsor states in summary that there was a statistically significant decrease in AST in HD males, but this is not discernible from the data. Increased cholesterol was observed in HD females and noted in sponsor summary, but this was not statistically significant and was not likely to be biologically significant. Decreased Cl- in females at L, M2 and HD groups (117, 111*, 112, 110*, 109*). These were within historical control levels.

Gross pathology: No treatment-related findings. Two females (M1 and M2) had fluid-filled uterine horns at terminal necropsy.

Toxicokinetics: B2036-PEG was systemically available after subcutaneous injection. Serum concentrations were greater after administration of 10 mg/kg than 0.3 mg/kg. (~25X). Mean serum concentrations were 192 and 4877 ng/ml for the 0.3 and 10 mg/kg doses, respectively. Thus bioavailability compared to IV injections appears to be low (~30-fold less than IV).

KEY STUDY FINDINGS:
There were no clear dose-related toxicities observed in this acute SC toxicity study in mice. Doses up to 10 mg/kg were well tolerated acutely. Based on surface area comparisons to the proposed daily dosing of 20 mg/day in humans, this represents a multiple of human exposure of ~ 3X. However, the bioavailability of SC injections compared to IV was quite low.

Study Title: 14-Day Intravenous Toxicity In CD-1 Mice
Study No: SEN-106
Amendment #034, Vol 3#, and page #: IND page numbers not provided.
Conducting laboratory and location:
Date of study initiation: January 11, 1996.

GLP compliance: Yes
QA- Report Yes (X) No ()

METHODS:
Dosing: 0, 0.1, 0.3, 1.0, and 3.0 mg/kg/day of B2036PEG via the tail vein. Animals had access to food and water *ad libitum* throughout the study and were housed individually after randomization. Animals for blood sampling were fasted 4 h prior to sampling.
species/strain: CD-1 mice.
#/sex/group or time point: 10/sex/group.
age: 6 weeks.
weight:23-32 g males, 19-27 g females.
satellite groups used for toxicokinetics or recovery: Three additional groups (2/sex/group control, 0.1 and 3 mg/kg/day) were used to measure plasma concentrations. Blood was collected at a single time point on day 14 of treatment either at pretest or 1 h post-dose from mice dosed at 0.1 or 3.0 mg/kg/day.
dosage groups in administered units: 0, 0.1, 0.3, 1.0, and 3.0 mg/kg/day
route, form, volume, and infusion rate: IV B2036PEG via the tail vein. 5 ml/kg.
Drug, lot#, radiolabel, and % purity: Lot number 23663-38.
Formulation/vehicle: Nutropin buffer.

OBSERVATIONS AND TIMES:
Clinical signs: Twice daily (morning and evening for mortality and moribund condition). Clinical signs were observed prior to and immediately following dosing.
Body weights: Day of randomization, days 0, 7, and 14 (fasted 4 h and prior to sacrifice on day 14).
Food consumption: Weekly on days 0, 7, 13.
Ophthalmoscopy: Prior to randomization then prior to terminal sacrifice.
Hematology: HGB, HCT, RBC, Total, differential WBC, Plt, MCV, MCH, MCHC
Clinical chemistry: Glu, Glob, Tbili, Cl, Ca, Na, CK, A/G ratio, Phos, Chol, Creat, Alb, K, TP, AST, ALT, BUN, ALP.
Organ weights: See Histopathology table in appendix
Gross pathology: Not specified.
Histopathology: See Histopathology table in appendix. Standard H&E staining evaluated control and HD only. Injection sites and liver were evaluated in all groups.
Toxicokinetics: Three additional groups (2/sex/group control, 0.1 and 3 mg/kg/day) were used to measure plasma concentrations. Blood was collected at a single time point on day 14 of treatment either at pretest or 1 h post-dose from mice dosed at 0.1 or 3.0 mg/kg/day.

RESULTS:

Clinical signs: No dose-related mortalities. One control was dead on day 2 of dosing with indications of trauma. All dose groups exhibited scabs at the injection sites with a higher incidence for males (L and M1 dose groups). Scabs were evident in males after the second dose; for females, scabs were noted after the seventh dose. There was only one control female that had this finding. One M1 male and one M2 female exhibited signs of hardness of the tail, edema and/or erythema occasionally during the study.

Body weights: Decrease in body weight gain in M2 and HD of both sexes. Statistically significant for males only. (Control, L, M1, M2, HD (grams):
♂: 3,2,2,1*,1** (*p<0.05, **p<0.01)
♀: 2,2,2,1,1
Overall body weights were not biologically different.

Food consumption: No treatment-related effects.

Ophthalmoscopy: Central corneal opacities: 1 control female, two LD females and one M2 male. Focal striate retinopathy unilaterally in one M2 female and one HD male. Partial retinal detachment unilaterally for one M2 female. All were considered unlikely to have been treatment-related since the findings were not dose-related.

Hematology: No clear treatment-related effects. Statistically significant increase for RBC (all doses), Hb (L, M1) and HCT (L, M1, M2) for treated males, but sponsor claims this appears likely due to the fact that the controls were slightly lower than normal. Comparison of controls for these parameters for the mouse studies reviewed in this document are shown in the table 2 below.

Table 1: Changes in Hematology Parameters During 14-Day Mouse Toxicity

DOSE (mg/kg) – PARAMETER ↓	*CONTROL*	*0.1*	*0.3*	*1.0*	*3.0*
RBC (10^6/mm^3)	8.54	9.53**	9.46**	9.34**	9.31**
HGB (g/dL)	13.9	15.3**	15.2**	14.7	14.7
HCT (%)	42.6	46.8**	46.5**	45.4*	45.1

Table 2: Control Values for Hematology Parameters from Mouse Studies in this Submission

		RBC 10^6/mm^3	HGB g/dL	HCT (%)
MALES	ACUTE STUDY #0406S35.001 IV	9.5	15.6	47.1
	ACUTE STUDY #0406S35.002 IV	9.08	14.8	44.3
	ACUTE STUDY #0416S35.001 SC	9.07	15.1	46.9
	14-DAY STUDY IV	8.54	13.9	42.6
FEMALES	ACUTE STUDY #0406S35.001 IV	9.2	15.5	46.6
	ACUTE STUDY #0406S35.002 IV	9.1	14.6	44.0
	ACUTE STUDY #0416S35.001 SC	9.45	15.5	46.4
	14-DAY STUDY IV	9.44	15.3	46.3

Clinical chemistry: Elevated total protein, albumin, calcium (M2 and HD of both sexes - Ca in females not statistically significant) and phosphorus (Males at M1, M2, and HD). Decreased glucose in all treated groups of both sexes was statistically significant except for M2 females. Decreased chloride was observed in M2 males and LD, M2 and HD females. All treated groups had statistically significant decreases in alkaline phosphatase although this was not significant for low dose males. Statistically significant lower BUN was observed in M1, M2 and HD males and in HD females.

Table 3: Summary Of Blood Chemistry Findings In Mice Treated 14 Days Intravenously

	MALES				
DOSE (mg/kg)→	CONTROL	0.1	0.3	1.0	3.0
PARAMETER↓					
TOTAL PROTEIN (g/dL)	5.4	5.6	5.8	6.7*	6.4*
ALBUMIN (g/dL)	3.8	4.1	4.3	5.0*	4.6*
CALCIUM (mg/dL)	9.6	9.6	9.6	10.2*	10.3**
PHOSPHORUS (mg/dL)	10.4	12.0*	11.8*	10.9	11.8*
GLUCOSE (mg/dL)	231	206*	176*	192*	201
CHLORIDE (mEq/L)	110	111	112	107*	108
BUN (mg/dL)	26	25	20*	18*	17*
ALP (IU/L)	104	81	54*	38*	40*
	FEMALES				
TOTAL PROTEIN (g/dL)	5.3	5.3	5.5	6.4*	6.7*
ALBUMIN (g/dL)	3.9	4.0	4.2	4.7*	5.0*
CALCIUM (mg/dL)	9.7	9.7	9.5	10.3	10.3
PHOSPHORUS (mg/dL)	10.4	10.7	10.8	10.5	11.1
GLUCOSE (mg/dL)	216	179*	168*	189*	171*
CHLORIDE (mEq/L)	117	114*	115	113*	113*
BUN (mg/dL)	19	16	17	18	15*
ALP (IU/L)	133	91*	60*	44*	33*

Urinalysis: No data.

Organ Weights: No clear dose-related changes observed. Decreased kidney weights (absolute (16%) and relative to body weight (15%)) in high dose males. Statistically significant increased liver: body weight ratio in high dose females (8%). Liver: body weight in M1 and M2 males was lower than controls (12-13%). There were no statistically significant findings in these tissues relative to brain. Slightly higher heart: body weight ratio in all groups of treated males (6,12,12,12%).

Gross pathology: Lesions (scabs) at injection sites noted in all groups.

Histopathology: M1 and M2 males and females had areas of centrilobular basophilia (Increased RER? - corresponds to increased total protein and albumin in M1 and M2 groups) and decreased hepatocellular pallor (decreased glycogen content? This would correspond to the decreased glucose levels noted) in all treatment groups. These were rated slight to mild. Decreased ALP attributed to decreased osteoclast activity, but this should be characterized further by analyzing the tissue source of ALP. Dose related findings at injection site include: local subacute cellulitis, edema, hemorrhage, epidermatitis, hyperkeratosis, subacute vasculitis or ulceration. Most of these findings were rated slight to mild. One M2 female had a severe large ulcerated lesion on the tail.

Toxicokinetics: B2036-PEG was systemically available after multiple IV injections. Serum concentrations were greater after administration of 3 mg/kg than 0.1 mg/kg. (~50X).

KEY STUDY FINDINGS:

1. Decrease in body weight gain in M2 and HD of both sexes. Statistically significant for males only. This might be expected for the biological activity of the test agent, but early studies suggested that the mouse was a poor responder to this agent. The cause of the loss of body weight gain is unclear. Overall bodyweight changes did not appear to be biologically important.
2. Elevated total protein, albumin, calcium (M2 and HD of both sexes - Ca in females not statistically significant) and phosphorus (Males at M1, M2, and HD). Decreased glucose in all treated groups of both sexes was statistically significant except for M2 females. Decreased chloride was observed in M2 males and LD, M2 and HD females. All treated groups had statistically significant decreases in alkaline phosphatase although this was not significant for low dose males. Statistically significant lower BUN was observed in M1, M2 and HD males and in HD females.

3. Decreased kidney weights (absolute (16%) and relative to body weight (15%)) in high dose males. Statistically significant increased liver: body weight ratio in high dose females (8%). Liver: body weight in M1 and M2 males was lower than controls (12-13%). There were no statistically significant findings in these tissues relative to brain. Slightly higher heart: body weight ratio in all groups of treated males (6,12,12,12%).
4. Decreased ALP attributed to decreased osteoclast activity, but this should be characterized further by analyzing the tissue source of ALP.
5. There were no clear dose-related toxicities observed in this 2-week IV toxicity study in mice although there were some relatively slight findings of decreased body weight gain, increased serum total protein and calcium and phosphorus, decreased glucose, Cl, and alkaline phosphatase. There were slight to mild findings of injection site reactions. Doses up to 3 mg/kg were well tolerated for 2 weeks. Based on surface area comparisons to the proposed daily dosing of 20 mg/day in humans, this represents a multiple of human exposure of ~ 1.4X.

Study Title: 14-Day Subcutaneous Toxicity In CD-1 Mice
Study No: SEN-107
Amendment #034, Vol #4, and page #: IND page numbers not provided.
Conducting laboratory and location:
Date of study initiation: January 18 (males) and 19 (females), 1996.
GLP compliance: Yes
QA- Report Yes (X) No ()

METHODS:
Dosing: 0, 0.1, 0.3, 1.0, and 3.0 mg/kg/day of B2036PEG SC as a bolus. Animals had access to food and water *ad libitum* throughout the study and were housed individually after randomization. Animals for blood sampling were fasted 4 h prior to sampling.
species/strain: CD-1 mice.
#/sex/group or time point: 10/sex/group.
age: 6-weeks
weight: 24-30 g males, 18-28 g females.
satellite groups used for toxicokinetics or recovery: Three additional groups (2/sex/group control, 0.1 and 3 mg/kg/day) were used to measure plasma concentrations. Blood was collected at a single time point on day 14 of treatment either at pretest or 1 h post-dose from mice dosed at 0.1 or 3.0 mg/kg/day.
dosage groups in administered units: 0, 0.1, 0.3, 1.0, and 3.0 mg/kg/day
route, form, volume, and infusion rate: SC 5 ml/kg.
Drug, lot#, radiolabel, and % purity: Lot number 23663-38.
Formulation/vehicle: Nutropin buffer.

OBSERVATIONS AND TIMES:

Clinical signs: Twice daily (morning and evening for mortality and moribund condition). Clinical signs were observed prior to and immediately following dosing.
Body weights: Day of randomization, days 0, 7, and 14 (fasted 4 h and prior to sacrifice on day 14).
Food consumption: Weekly on days 0, 7, 13.
Ophthalmoscopy: Prior to randomization then prior to terminal sacrifice.
Hematology: HGB, HCT, RBC, Total, differential WBC, Plt, MCV, MCH, MCHC
Clinical chemistry: Glu, Glob, Tbili, Cl, Ca, Na, CK, A/G ratio, Phos, Chol, Creat, Alb, K, TP, AST, ALT, BUN, ALP.
Organ weights: See Histopathology table in appendix
Gross pathology: not specified.

Histopathology: See Histopathology table in appendix. Standard H&E staining evaluated control and HD only. Injection sites and liver were evaluated in all groups.
Toxicokinetics: Three additional groups (2/sex/group control, 0.1 and 3 mg/kg/day) were used to measure plasma concentrations. Blood was collected at a single time point on day 14 of treatment either at pretest or 1 h post-dose from mice dosed at 0.1 or 3.0 mg/kg/day.

RESULTS:

Clinical signs: No treatment-related signs were observed. There were no dose-related mortalities. A single low dose female was found dead on day 9 of dosing. There was a dark discoloration of the skull and brain consistent with accidental trauma.
Body weights: No treatment-related effects. There was a statistically significant increase in the body weight gain of one low dose female which was considered incidental to treatment. Overall body weights were not biologically different.

Food consumption: No treatment-related effects.

Ophthalmoscopy: No treatment-related lesions were noted.

Hematology: No treatment-related effects. Statistically significant decrease in Hb (5%), HCT (5%) and increase in platelet count values (917 in control, 1248 $10^3/mm^3$ in high dose) for M2 females. However, there was a decrease in platelet count in low dose males. These findings are incidental.

Clinical chemistry: Elevated total protein (both sexes: M2 and HD), albumin (both sexes M1, M2, HD), calcium (both sexes at M2 and HD), cholesterol (HD males, M2 and HD females) and albumin/globulin ratio (all treated male groups). Decreased glucose (M2, HD males, M2 females), potassium (M2 females), chloride ((HD males, M2 and HD females) and ALP (M2, HD males, M1, M2,HD females). Globulin varied: Increased in M2 and HD females and decreased in LD and M1 males.

Table 4: Summary Of Blood Chemistry Findings In Mice Treated 14 Days Subcutaneously

		MALES			
DOSE (mg/kg)– PARAMETER:	CONTROL	0.1	0.3	1.0	3.0
TOTAL PROTEIN (g/dL)	5.5	5.5	5.7	6.9**	7.8**
ALBUMIN (g/dL)	3.7	4.0	4.1*	5.0**	5.7**
GLOBULIN (g/dL)	1.8	1.5**	1.6*	1.9	2.0
A/G RATIO	2.0	2.8**	2.7**	2.7**	2.9**
CALCIUM (mg/dL)	9.9	9.7	9.9	10.7**	11.3**
POTASSIUM (mEq/l)	10.5	10.1	9.9	9.8	9.5
GLUCOSE (mg/dL)	234	222	213	196**	188**
CHLORIDE (mEq/L)	113	115	115	112	110*
CHOLESTEROL (mg/dL)	140	134	131	175	222**
ALP (IU/L)	80	84	76	30**	23**
		FEMALES			
TOTAL PROTEIN (g/dL)	5.4	5.2	5.8	7.2**	7.4**
ALBUMIN (g/dL)	4.1	4.0	4.5*	5.6**	5.7**
GLOBULIN (g/dL)	1.3	1.2	1.3	1.6**	1.7**
A/G RATIO	3.2	3.2	3.5	3.4	3.4
CALCIUM (mg/dL)	9.9	9.7	10.2	11.0**	11.2**
POTASSIUM (mEq/l)	9.6	9.2	9.6	8.4**	8.9
GLUCOSE (mg/dL)	192	170	186	165	172
CHLORIDE (mEq/L)	116	114	115	112**	112**
CHOLESTEROL (mg/dL)	108	94	106	153**	158**
ALP (IU/L)	90	79	54*	23**	19**

Organ Weights: No clear dose-related changes observed. In M1 males, there was a statistically significant decrease (14%) in liver: body weight ratio.

Gross pathology: Lesions at injection sites noted in some animals (1 M2 male; 1 control, 1 LD, 1 M2 and 1 HD female), however, these seemed to be related to injection procedures, not drug. Severity ratings were minimal to mild.

Histopathology: M2 and high dose females and males had areas of centrilobular increased hepatocellular basophilia (M2 and HD of both sexes. 8/10 in both male groups; 10/10 ,9/10 in female groups. Increased RER?) and decreased hepatocellular pallor (Males: 10/10 in control, LD and M1, 2/10 in M2 and HD. Females: 10/10 in control, LD and M1, 0/10 in M2 and 1/10 in HD. Decreased glycogen content). These were rated in severity from minimal to slight/mild.

The sponsor attributes many findings to physiological effects rather than toxicological effects of the drug. However, isn't this agent inactive in mice?

Toxicokinetics: Data were not presented. Consultant suggests that doses are approximately 12-fold greater at 3 compared to 0.1 mg/kg. However, the numbers quoted for plasma levels (667 and 236 ng/ml for 0.01 and 3.0 mg/kg, respectively) do not support this. Without the data, these numbers cannot be relied upon. **Note added in proof: sponsor corrected values to be 667 and 8236 ng/ml for the 0.1 and 3 mg/kg doses. (fax of 4/22/99)**

KEY STUDY FINDINGS:

1. There were no treatment-related effects on body weight, food consumption, ophthalmoscopy or hematology.
2. Elevated total protein (both sexes: M2 and HD) albumin (both sexes M1, M2, HD and calcium (both sexes at M2 and HD) were consistent with the IV study. Elevated cholesterol (HD males, M2 and HD females) and albumin/globulin ratio (all treated male groups) were noted in this study. Decreased glucose (M2, HD males, M2 females), potassium (M2 females), chloride ((HD males, M2 and HD females) and ALP (M2, HD males, M1, M2,HD females)were observed in this study as well and were consistent with the IV study. Globulin varied: Increased in M2 and HD females and decreased in LD and M1 males.
3. There were no significant treatment-related effects on organ weights.
4. Local effects at the injection site did not appear to be dose related.
5. Decreased ALP attributed to decreased osteoclast activity, but this should be characterized further by analyzing the tissue source of ALP.
6. There were minimal to mild findings of basophila in the liver, which appeared dose related, but did not appear to correlate with specific liver toxicity (no transaminase elevations or necrosis). This could be related to the changes in glucose/glycogen levels noted.
7. There were no clear dose-related toxicities observed in this 2-week IV toxicity study in mice There were some relatively slight findings of decreased body weight gain, increased serum total protein and calcium and phosphorus, decreased glucose, Cl, and alkaline phosphatase. There were slight to mild findings of injection site reactions. Doses up to 3 mg/kg were well tolerated for 2 weeks. Based on surface area comparisons to the proposed daily dosing of 20 mg/day in humans, this represents a multiple of human exposure of ~ 1.4X.

OVERALL TOXICOLOGY SUMMARY:

Toxicological findings were slight and tended to be inconsistent between experiments within the same species as well as between species. This may be a reflection of the low multiples of exposure. However, there were some hematologic and blood chemistry effects that were consistent. These slight changes were detected in both species even though mice were supposedly not physiologically responsive to the growth hormone agonist:
Decreased Hb, RBC, some decrease in APTT and a decrease in ALP (type not specified). A slight decrease in body weight was noted in some mouse experiments and in monkeys. While this might be expected in monkeys due to the activity of the compound, it is not clear why this occurred in

mice, which are presumably unresponsive. Also noted were increase in total protein and albumin. There were slight injection site reactions noted in both species.

SPECIAL TOXICOLOGY STUDIES:

General Comments: This study is a new submission to the IND. It characterizes the RIA used to quantitate B2036-PEG.

Study Title: The quantitative determination of B2036-PEG in mouse serum by RIA (Validation of RIA)

Study No: SEN-002
Amendment #034, Vol #1, and page #: (IND page numbers not provided)
Conducting laboratory and location:

Date of study initiation: Not provided Study dated 8/1/97, Signed by Sensus consultant 12/16/98.
GLP compliance: Yes
QA- Reports Yes (X) No ( ):

METHODS: Standard RIA using mouse serum, Sheep Anti-Rabbit IgG precipitating Ab and Rabbit Anti-Mouse specific antibody from Standards were diluted in RIA buffer. "Samples" were mouse serum spiked with B2036-PEG ranging from 4.8 –300 ng/ml.
Drug, lot#, radiolabel (if applicable), and % purity:
22938-45 (Genentech, So. San Francisco, CA) Purity 93.5% (correction factor used in RIA calculations).
003007 Purity 100% (no correction factor used).
[^{125}I]-B2036-PEG was synthesized with the chloramine T method.
Formulation/vehicle: mouse serum,
160296.

Key finding(s):
1. B2036-PEG was quantitated in mouse serum to validate the RIA. Antibody was Rabbit-anti-mouse produced by
2. LLOQ ~
3. LLOD ~
4. ULOQ~
5. Linearity of response was demonstrated for the entire range used.
6. Reproducibility of standard curve was between 1 and 6.8% based on comparisons over the range of the curve. Coefficient of variation (n=3) at the mid region of the curve was 21.1%.
7. Accuracy of the curve was based on LLOQ-QC, LQC, MQC, HQC and ULOQ-QC. % Nominal values were respectively.
8. Interassay variability ranged between 3.5-17% depending upon the point on the curve evaluated.
9. Cross reactivity: 100% for B2036, 22% for B2036-PEG and 23% for hGH.
10. Cross reactivity to mouse GH could not be evaluated due to unavailability of the purified mouse hormone. However, LLOQ in 10 lots of mouse serum was lower than the LLOQ of the assay of indicating the absence of cross-reactivity of mouse GH with the antiserum. It does not cross react with human prolactin or human placental lactogen.
11. B2036-PEG was stable in mouse serum at room temperature for at least h. and for freeze-thaw cycles. B2036-PEG was stable in mouse serum for at least days at when compared against freshly prepared control samples.

Study Title: ACTION OF GROWTH HORMONE ANTAGONISTS IN PRIMATES
Study No: SEN-102
Amendment #0034, Vol 1 #, and page #: Pages not numbered.
Conducting laboratory and location: Sponsored by Sensus Corporation.

Date of study: September 4, 1997.
GLP compliance: No
QA- Reports Yes ( ) No (X):

METHODS:
Four studies were conducted on captive born adolescent (2-4yrs/weight 3.5-6 kg) male rhesus monkeys to determine the effects of acute administration of growth hormone antagonists (G120K, B2024, B2024-PEG, B2036, B2036-PEG) on physiological parameters (IGF-1, IGFBP3, and the antigenicity of GHA's).

Captive born adolescent (2-4yrs/weight 3.5-6 kg) male rhesus monkeys were used in these studies. Water was available *ad libitum*. Food was provided prior to 0900h and between 1500 and 1600 h. Amounts provided were sufficient for the animal to consume as much as desired within the feeding periods. Serum IGF-1 was determined following removal of binding proteins. RIA sensitivity was 10 ng/ml for IGF-1. IGFBP-3 was determined by IRMA with commercially available kits which was validated for monkey serum. Lower limit of detection was Serum insulin was measured by commercially available RIA kit (sensitivity was 5U/ml). GH was measured by ELISA by Dr.

Study 1: PK and PD of GH Antagonists: 5 monkeys received, in random order, placebo, 1 mg/kg B2036-PEG both IV and SC. (10 mg/ml concentration dosing solution). Serum samples were taken at 0, 1, 5, 10,15 and 30 min after dosing and 1,2,4,8 h after dosing and 1,2,3,4,5,6,7 and 14 days after dosing. The second treatment commenced on day 14 after the first injection and the third study began 21 days after the second injection.

Study 2: Dose Response Characteristics of G120K-PEG and B2036-PEG: 12 additional monkeys were dosed with G120K-PEG or B2036-PEG at 0.03, 0.10, 0.30 and 1.0 mg/kg. (Dose volume 0.4-0.8 ml). Data from study 1 were included in the analysis to increase sample size. This was a crossover design with dosing in two separate treatments/animal. "Wash out" period was two weeks.

Study 3: Effect of Repeated Administration of Varying Doses of B2036-PEG. 16 male monkeys, not used previously, 4/group received placebo, B2036-PEG at 0.1, 0.3, and 1.0 mg/kg in 6 weekly injections (total 7 injections). Samples were obtained prior to each injection, then once daily up to 7 days post dosing. Samples were analyzed for IGF-1, insulin, clinical chemistry (including glycosylated hemoglobin) and lipids.

Study 4: Assessment of antigenicity of GHA's: 12 monkeys not previously treated were randomly assigned to 1 of 6 groups (n=2) which received one of the 6 GHA's daily at 1.0 mg (~0.2 mg/kg and 0.15 mg/kg for the two HD monkeys) SC for 2 weeks. Blood samples were collected predosing, and weekly for weeks after initial dose. These samples were analyzed for IGF-1, IGFBP-3 and antigenicity assessment.

Drug, lot#, radiolabel (if applicable), and % purity: Genentech was source of GHA's, lot numbers not provided.
Formulation/vehicle: not specified.

RESULTS:

I. PK/PD of single IV or SC dose of B2036-PEG: Serum IGF-1 and IGFBP-3 were lowered within the first 4 h postdose by either route. This continued to decline, reaching a nadir at 4 days (SC = -85.4 % ±1.0; IV =-85.8% ± 0.7. Levels remained suppressed through day 7, then rose to near baseline by day 14 and stabilized thereafter. It appeared that IGFBP-3 concentrations declined more rapidly after IV administration compared with SC. Nadir levels on day 4 (SC =-38.3% ± 2.2; IV = -41.4% ± 2.4).

II. Dose response of G120K-PEG and B2036-PEG: B2036-PEG appeared to be more effective than G120K-PEG at lowering IGF-1 and IGFBP-3. Effect was dose dependent and evident at the two higher doses. Profiles of IGF-1 and IGFBP-3 changes were similar to the above results. G120K-PEG did not appear to have an effect on IGFBP-3 at any dose.

III. Multiple-Dose/Dose Response with B2036-PEG: High dose only was able to suppress IGF-1 through 7 days following treatment. As in the previous study, 0.3 mg could suppress IGF-1 by day 3, but not through day 7. Repeated weekly injections of this dose was not able to suppress IGF-1 through day 7. Little effect was noted in the 0.1 and 0.3 mg/kg doses. In contrast, the high dose (1.0mg/kg) decreased IGF-1 levels by week 1 and appeared to decline progressively through 7 weeks. Effect at week 1 was not as dramatic (only a 38% decrease in study III vs 84% decrease in study I), but this was attributed to a lower baseline in study III. Preliminary results on GH suggest that there may be an inverse relationship between serum GH and the degree to which IGF-1 is suppressed. (Current studies are assessing the cross-reactivity of B2036-PEG with the monoclonal antibody to hGH used in the ELISA to rule out that the elevated GH estimates are due to binding of the GHA to the antibody complex). There appeared to be no metabolic consequences to the apparent increased serum GH (insulin, glucose and total cholesterol were examined).

IV. Assessment of antigenicity: B2036-PEG suppressed IGF-1 levels while the other 5 GHA's tested in this study did not. A similar pattern was observed for serum IGFBP-3. (dose of 1 mg daily corresponded to 0.21 and 0.15 mg/kg for the high dose animals. Total dose received over the 7 day period was approximately 1.3 mg/kg). Data on antigenicity indicate that one of the 2 monkeys administered B2036 developed a weak positive response of 12% specific binding on day 28 with a titre of 1:220.

Key finding(s):

1. Serum IGF-1 and IGFBP-3 were lowered within the first 4 h after a single dose by either IV or SC route. This continued to decline, reaching a nadir at 4 days (SC = -85.4 % ±1.0; IV =-85.8% ± 0.7. Levels remained suppressed through day 7, then rose to near baseline by day 14 and stabilized thereafter. It appeared that IGFBP-3 concentrations declined more rapidly after IV administration compared with SC. Nadir levels on day 4 (SC =-38.3% ± 2.2; IV = -41.4% ± 2.4).
2. Profiles of IGF-1 and IGFBP-3 changes were similar. Effect was dose dependent and evident at the two higher doses.
3. High dose only was able to suppress IGF-1 through 7 days following treatment. As in the previous study, 0.3 mg could suppress IGF-1 by day 3, but not through day 7. Repeated weekly injections of this dose was not able to suppress IGF-1 through day 7. Little effect was noted in the 0.1 and 0.3 mg/kg doses. In contrast, the high dose (1.0mg/kg) decreased IGF-1 levels by week 1 and appeared to decline progressively through 7 weeks. Effect at week 1 was not as dramatic (only a 38% decrease in study III vs 84% decrease in study I), but this was attributed to a lower baseline in study III.
4. Assessment of antigenicity: B2036-PEG suppressed IGF-1. A similar pattern was observed for serum IGFBP-3. (dose of 1 mg daily corresponded to 0.21 and 0.15 mg/kg for the high dose animals. Total dose received over the 7 day period was approximately 1.3 mg/kg). Data on

antigenicity indicate that one of the 2 monkeys administered B2036 developed a weak positive response of 12% specific binding on day 28 with a titre of 1:220

Study Title: **Interaction Between Somavert® and Regular or NPI Insulin**
Study No: SEN-203.
Amendment #034, Vol #4, and page #: IND page numbers not specified.
Conducting laboratory and location:

Date of study initiation: Letter describing results dated January 27, 1998.
GLP compliance: NO
QA- Reports Yes ( ) No (X):

METHODS:
This was a preliminary non-GLP study to determine if there was an interaction between Somavert® and regular or NPI insulin.

Samples of mixtures of Somavert® and either NPI insulin or regular insulin were stored at 5 or 25°C for various times up to 3 days

Observations and times: After storage for up to 3 days, samples were submitted for UV scanning to determine changes in UV absorbance.
Results: No significant changes were detected for UV absorbance when the mixtures were stored up to 3 days.

OVERALL SUMMARY AND EVALUATION:

Introduction:
Pre-IND meeting held with sponsor January 24, 1996. Alex Jordan and Ron Steigerwalt were pharmacologists present. Key toxicology items discussed: Dr. Jordan indicated that the monkey studies would probably be sufficient and also noted that carcinogenicity studies would eventually be required. Teratology should be performed in a species in which the drug works. There was a meeting with the sponsor regarding preclinical requirements for both acromegaly indications. The CDER reproductive toxicology committee was consulted as to the appropriate design of studies. Joe DeGeorge was consulted regarding carcinogenicity studies. For Acromegaly, the Reproductive Toxicology committee recommended a modified Segment I/Segment II approach. The results of these consults were incorporated into a report of the minutes of the May 22, 1998 meeting (see copy of a summary of conclusions from the minutes from May 22, 1998 meeting with the sponsor. These comments were communicated to the sponsor in a fax on September 17, 1998.

The present submission provides some additional studies with validation of the RIA for B2036-PEG. Final study reports are provided for some of the studies that were initially reviewed under the initial submission. Additional PK data from the toxicology studies was provided. These reviews are reproduced in this review with the PK data appended.

Safety Evaluation: The sponsor has switched from the initially proposed weekly dosing to daily dosing. The exposure for daily dosing in humans on a weekly basis exceeds the single dose/week initially proposed by nearly 2-fold the highest exposure in the animal studies. The highest exposure in the animal studies approximates the proposed human exposure. Toxicity findings appeared to be relatively minor and there were no kidney findings in the multiple daily dose studies in mice or monkeys (see original review) which would suggest that there is not a problem with kidney effects noted in animal studies with a PEGylated TNF. The sponsor has been allowed to proceed in human trials without specific coverage of daily dosing in animal studies of equal

duration. This was communicated to the sponsor in a fax. The specific points are reproduced after the conclusions section of this document.

Clinical Relevance of Safety Issues: No significant safety issues are evident. However, sponsor is dosing humans daily beyond duration covered by animal toxicity studies. Highest exposures in animal toxicology studies approximate the human exposure when calculated on a mg/m^2 basis. It is not clear what actual plasma level multiples would be.

Conclusions: The studies presented are final reports of studies already reviewed. This submission does not add significantly to the overall understanding of the toxicity of this product. The sponsor basically dismisses some hematological findings that nonetheless were consistent between mouse and monkey studies. These were not severe toxic findings, and can be monitored in clinical trials.

STUDIES REQUESTED FOR B2036-PEG (FROM MEETING MINUTES OF MAY 22 1998 MEETING-MINUTES FINALIZED AUGUST 10,1998 AFTER COMMITTEE CONSULTATIONS) THESE HAVE BEEN COMMUNICATED TO THE SPONSOR

The following provides an outline for the preclinical studies necessary for filing of an NDA for Trovert:

1. Toxicity studies: A three-month toxicity study in rats with daily dosing and a minimum high dose of 10-fold (preferably 25-fold) the human maximum AUC will be provided for the filing of an acromegaly indication.

2. Reproductive toxicity: For the acromegaly indication, the use of the rabbit as a single species in the reproductive toxicity studies should suffice. However, the Reproductive Toxicology Committee recommended that a modified Seg I/Seg II approach be undertaken, in which one set of experiments begins treatment of inseminated does on the day of insemination, and continues treatment through Day 7; and the second set of experiments treats during the traditional organogenesis period (Days 6-19).
identify a second responsive species for further reproductive toxicology testing. In particular, the committee felt that more effort should go into validating (or ruling out) the mouse as a model. The Division concurs with the committee recommendations.

3. Carcinogenicity studies: The sponsor should provide the following at the NDA submission for acromegaly: GH binding studies as outlined at the May 22 meeting, and the PC-3 tumor cell transplant study proposed by the sponsor during the 6/8/98 teleconference. A single carcinogenicity study submitted Phase IV could cover the acromegaly indication.

It should be understood that the above recommendations are based on the proposal for daily clinical dosing with Trovert™ at 20 mg/kg/day. Changes in the clinical proposal or formulation could necessitate additional preclinical studies.

RECOMMENDATIONS:

Internal comments: The requirements for toxicology studies to support NDA's for both the acromegaly [redacted] indications have been communicated to the sponsor (see copy of items above). No additional preclinical data have been submitted since this communication.

External Recommendations (to sponsor): none.
Future development or NDA issues: Daily dosing for 3 months as described in the attached fax should help determine if there are any significant toxicities with this product.

DRAFT LETTER CONTENT FOR SPONSOR:

1. According to the cover letter for amendment #034 dated January 25, 1999, The final report for SEN-109 (the 26-week monkey study) is referenced as being included in this amendment. Please be advised that this study was not included. Since this study has been submitted in draft form and entails weekly dosing which is no longer relevant for human exposure, it is not critical to submit the final study at this time unless significant changes have occurred in the finalization of the report.
2. In the summary provided for SEN-107, reference is made to toxicokinetic data from a PK analysis study SEN-004. Have these data been submitted? The plasma levels quoted in the summary (667 and 236 ng/ml for the 0.1 and 3.0 mg/kg doses, respectively) do not appear to be correct. This summary also suggests that the difference in serum concentrations is ~ 12-fold, which would not be evident from the reported plasma levels.

Reviewer signature/team leader signature [Concurrence/Non-concurrence]

Ronald W. Steigerwalt, Ph.D.
Pharmacology Team Leader

cc: IND Arch
HFD510
HFD510/Steigerwalt/CKing/Perlstein
Review Code: DE
Filename: [redacted]

Appendix/attachments:
1. Histopathology table
2. Fax to sponsor outlining studies required to support an NDA for acromegaly.

Addendum 1:Histopathology Inventory for IND #

Study	SEN-106 14 day IV	SEN-107 14 day SC
Species	*Mouse*	*Mouse*
Adrenals	X*	X*
Aorta	X	X
Bone Marrow smear		
Bone (femur)	X	X
Brain	X*	X*
Cecum	X	X
Cervix		
Colon	X	X
Duodenum	X	X
Epididymis	X	X
Esophagus	X	X
Eye	X	X
Fallopian tube		
Gall bladder	X	X
Gross lesions	X	X
Harderian gland		
Heart	X*	X*
Hyphophysis		
Ileum	X	X
Injection site	X	X
Jejunum	X	X
Kidneys	X*	X*
Lachrymal gland	X	X
Larynx		
Liver	X*	X*
Lungs	X	X
Lymph nodes, cervical		
Lymph nodes mandibular		
Lymph nodes, mesenteric	X	X
Mammary Gland	X	X
Nasal cavity		
Optic nerves	X	X
Ovaries	X*	X*
Pancreas	X	X
Parathyroid	X	X
Peripheral nerve		
Pharynx		
Pituitary	X	X
Prostate	X	X
Rectum	X	X
Salivary gland	X	X
Sciatic nerve	X	X
Seminal vesicles	X	X
Skeletal muscle	X	X
Skin	X	X
Spinal cord	X	X
Spleen	X*	X*
Sternum	X	X
Stomach	X	X
Testes	X*	X*
Thymus	X	X
Thyroid	X	X
Tongue	X	X
Trachea	X	X
Urinary bladder	X	X
Uterus	X	X
Vagina	X	X
Zymbal gland		

* organ weight obtained

REVIEW AND EVALUATION OF PHARMACOLOGY/TOXICOLOGY DATA:

KEY WORDS: 28 day Toxicology, daily dosing, Rhesus monkeys, Mouse PK data

Reviewer Name: Ronald W. Steigerwalt, Ph.D. Pharmacology Team Leader
Division Name: Division of Metabolic and Endocrine Drug Products (DMEDP)
HFD#510
Review Completion Date: March 22, 1999
Review number: 6

IND/NDA NUMBER: IND ——
Serial number/date/type of submission: SN#037, March 8, 1999 IT
Information to sponsor: Yes () No (X)
Sponsor (or agent): Sensus Drug Development Corp.; Austin, TX 78701

DRUG

Code Name: B2036-PEG
Generic Name: rhGH antagonist (PEGylated)
Trade Name: Trovert™
Molecular Formula/ Molecular Weight: B2036-PEG is a 191 amino acid polypeptide with two disulfide bonds. As an analogue of hGH, it contains nine substitutions in the first and third α-helices. There are 4-5 PEG moieties (~5000 Da each) covalently bound to amino groups on the surface of the molecules. This increases its mass to 42-46,000Da.

Relevant INDs/NDAs/DMFs: IND ——

Drug Class: Modified recombinant human growth hormone (antagonist).

Indication: Acromegaly

Clinical formulation: Per vial:
10 mg B2036 protein
36 mg Mannitol
1.36 mg glycine
—— sodium phosphate, pH 7.4.
To be reconstituted in water up to 10 mg/ml (for clinical testing).

Route of administration: SC daily

Proposed clinical protocol or Use: No new protocols proposed in this submission. Initial studies attempted weekly administration of 80 mg. This has been increased to daily administration of 20 mg/day (total 140 mg/week). Individual unresponsive patients have received higher doses on a daily basis.

Previous clinical experience: Phase I: Placebo controlled, double blind, single rising dose study in healthy male volunteers was "nearing completion" at time of submission. B2036-PEG was "well-tolerated at doses of up to 1.0 mg/kg and efficacious in suppressing IGF-1 concentrations" at 0.3 mg/kg (3/6 pts) and 1.0 mg/kg (6/6 pts). No serious AE's; Other AE's resolved without sequelae. PK appeared to be linear within the 0.03 and 0.1 mg/kg groups. The PK were non-linear at effective doses of 0.3 and 1.0 mg/kg.

A single dose study in six acromegalic patients was initiated at [redacted] The [redacted] No data are available at this time.

Additional studies have assessed the daily administration of **20 mg/day** in Acromegalic patients.

AE's: Preliminary data available:

0.5 mg/kg: (total 3 AE's)
1 subject unifocal ventricular extrasystoles before and after dosing. Extrasystoles remained 7 days later. Unlikely relationship with drug.

1 subject Abdominal pain and diarrhea (mild) Listed as remotely related to drug.
1 subject Headache (moderate). Listed as possibly related to drug.

0.1 mg/kg: 9 AE's: All mild: GI fullness, chest pressure, decreased appetite and headache considered remotely related to drug. Abdominal discomfort (possibly related) and nasal congestion and abnormal hunger were considered not related.

0.3 mg/kg: 3 AE's in 1 subject: rhinitis (day -3), oral herpes lesion (day -1). Mild itching at injection site lasting 12 h probably related to drug.

1.0mg/kg: 14 AE's: moderate epistaxis after minor trauma (remotely related) and headache (not related) 2 headaches were considered possibly related and parathesias of the tongue (2). Dry throat (2) and abdominal pain (1) were considered remotely related.

Phase 2: Protocol # SEN-3611: A Six Week, Double Blind, Placebo Controlled, Multiple Dose, Phase IIb study of B2036-PEG in the Treatment of Acromegaly. 45 patients (15/group) will receive weekly subcutaneous administrations for 6 weeks. Dose groups listed in protocol are 0, 30 and 80 mg of B2036-PEG. (Elsewhere in the submission, the dose selection is described as depending upon previous human experience). This was the initial plan, but the sponsor has since switched to daily dosing of 20 mg, with the possibility of increasing the dose further depending upon the patient response.

Disclaimer – use of sponsor's material: No sponsor material is reproduced in this report.

INTRODUCTION AND DRUG HISTORY:
Pre-IND meeting held with sponsor January 24, 1996. Alex Jordan and Ron Steigerwalt were pharmacologists present. Key toxicology items discussed: Dr. Jordan indicated that the monkey studies would probably be sufficient and also noted that carcinogenicity studies would eventually be required. Teratology should be performed in a species in which the drug works. There was a meeting with the sponsor regarding preclinical requirements for both acromegaly [redacted] The CDER reproductive toxicology committee was consulted as to the appropriate design of studies. Joe DeGeorge was consulted regarding carcinogenicity studies. For Acromegaly, the Reproductive Toxicology committee recommended a modified Segment I/Segment II approach. The results of these consults were incorporated into a report of the minutes of the May 22, 1998 meeting (see copy of a summary of conclusions from the minutes from May 22, 1998 meeting with the sponsor. These comments were communicated to the sponsor in a fax on September 17, 1998.

The present submission provides the finalized report for the 28 day monkey toxicology study with daily dosing. An interim report was reviewed in the initial submission. In addition, Some PK data

were submitted under SEN-004 and a dermal irritation study in rabbits is also provided (SEN-115). These latter 2 studies were not previously reviewed.

Studies reviewed within this submission:

1. SEN-108: B2036-PEG 28-Day Subcutaneous Toxicity Study in the Rhesus Monkey (With Every Other Day Dosing) (Toxicology section previously submitted and reviewed under SN#000). Page 3
2. SEN-115: Dermal Irritation Potential of B2036-PEG after SC Injection and Effect on Serum IGF-1 Levels in the Rabbit. Page 5

3. Studies not reviewed within this submission: SEN-004: Quantitative Determination of B2035-PEG in Mouse Serum by Radioimmunoassay. (final report, draft previously reviewed under SN#000). (No interpretable information provided in this report).

TOXICOLOGY:

General Comments: The 28 day monkey study was initially reviewed in SN#000. There was a statement of GLP compliance and signed QAU statement, indicating that the report was complete. This re-submission contains an additional integrated summary and PK data obtained during the initial study. **Note that dosing is every other day.**

Study Title: B2036-PEG 28-Day Subcutaneous Toxicity Study in the Rhesus Monkey with Every Other Day Dosing
Study No: SEN 108
Amendment #037, Vol #2, and page #: Pagination begins with 001 in second volume.
Conducting laboratory and location:
Date of study initiation: January 9, 1996.
GLP compliance: Yes
QA- Report Yes (X) No ()

METHODS:
Dosing: B2036PEG subcutaneously at 0, 0.1, 0.3, 1.0 and 3.0 mg/kg.
species/strain: Purpose-bred Rhesus monkeys from
#/sex/group or time point: 3/sex/group.
age: 30-34 months.
weight: 3.1-4.6 kg males, 2.4-3.9 females.
satellite groups used for toxicokinetics or recovery: No separate groups. Samples taken from test groups.
dosage groups in administered units: 0, 0.1, 0.3, 1.0 and 3.0 mg/kg SC administered every other day. There were a total of 14 dose administrations on days 1, 3, 5, 7, 9, 11, 13, 15, 17, 19, 21, 23, 25 and 27.

DOSING:
route, form, volume, and infusion rate: Dose volume was 0.5 ml/kg SC.
Drug, lot#, radiolabel, and % purity: Batch numbers 23663-38 and 23663-59.
Formulation/vehicle: Neutropin buffer.

OBSERVATIONS AND TIMES:
Clinical signs: Not specified.
Body weights: Weekly.
Food consumption: Measured daily, reported as weekly mean of g/animal/day

Ophthalmoscopy: Pretest, then week 4.
Hematology: terminal
Clinical chemistry: terminal
Urinalysis: terminal
Gross pathology: terminal
Organ weights: terminal
Histopathology: terminal
Toxicokinetics: All animals in groups 2 and 5 (0.1 & 3 mg/kg) pretest and on day 27 1 h post-dose.

RESULTS:
Clinical signs: No mortalities. Slight swelling at the injection site in some treated animals near the end of the treatment period. Not dose related.

Body weights: Small decreases in M2 and HD males (-15 and -12%, respectively). However, it is noted that in these two groups, the starting weights were lower than the other groups. Body weight curves were, in general, parallel.

Food consumption: No treatment-related effects.

Ophthalmoscopy: No treatment-related effects.

Hematology: HD females had lower Hb (12%), PCV (13%), RBC (16%). Actual values were within historical controls, but the sponsor stated that they did not rule out a treatment-related effect in this case. Similar trends were noted in males, but the extent was only 10% for Hb and 7-8% for RBC and PVC, respectively.

Clinical chemistry: M2 and HD animals tended to have lower ALP. Males: (IU/L): 1900, 1387, 1008, 723, 829. Females: 1485, 1484, 1260, 1315, 1225. Other changes were considered incidental. Slightly shorter APTT in M2, HD males (21.4, 21.1, 20.7, 19.8, 19.9). Also shortened in M1 and HD females (21.3, 20.5, 17.6, 20.6, 18.2).

Urinalysis: No treatment-related effects.

Organ Weights: No treatment-related effects.

Gross pathology: Higher incidence of swelling at the injection sites in treated animals. Incidence was not dose-related.

Histopathology: Minimal to slight hemorrhage at the injection site, although incidence across groups was not dose-related.

Toxicokinetics:

KEY STUDY FINDINGS:
1. This study was originally reviewed under the initial IND submission. The initial review is presented in the new pharmacology review format, but is basically unchanged from the initial review except for the addition of toxicokinetic data. This copy of the report is missing a number of pages throughout the toxicology section. The pagination is confusing, so it is difficult to specify all the pages that are missing. There is no clear pattern to the missing pages, so it is not possible to determine if this is due to a copying error. It is not possible to re-check data against the original review without these pages.

2. Hematology: Decreased Hb, PCV, RBC in HD females; Decreased APTT M2, HD males, M1, HD females
3. Blood Chemistry: Decreased ALP both sexes (type not specified)
4. There were no clear dose related findings in gross or histopathology examinations, although there was a treatment related, but not dose related occurrence of swelling at the injection site.
5. Note: there is a limitation in the toxicokinetic analysis since B2036-PEG cannot be distinguished from hGH or monkey GH. Since data were obtained only from the final dosing, it cannot be determined whether B2036-PEG accumulates with multiple dosing on this short-term basis. Data provided are of limited value.

SPECIAL TOXICOLOGY STUDIES:

Study Title: **Dermal Irritation Study of B2036-PEG after Subcutaneous Injection and Effect of B2036-PEG on IGF-1 levels in the Rabbit.**
Study No: SEN 115
Amendment #037, Vol #2, and page #: Not provided.
Conducting laboratory and location:
(Blood samples assayed for IGF-1 at
Date of study initiation: April 21, 1998
GLP compliance: No
QA- Reports Yes ( ) No (X):

METHODS:
DOSING: SC injection to 3 female rabbits/group of vehicle (not specified) and 3 mg/kg B2036-PEG at 0.15 ml/kg (3/group).
Drug, lot#, radiolabel (if applicable), and % purity: 362053A
Formulation/vehicle: Not provided.
Observations and times: Samples for determination of IGF-1 were taken predose, then on days 2, 3, 8 and 9 at 2 and 6 hours after dosing.
Results:
No deaths or changes in body weights were noted at termination. There were no drug-related histopathologic changes noted at the injection site other than irritation attributable to the injection itself.
IGF-1 kit was used to determine IGF-1 levels. B2036-PEG clearly reduced serum IGF-1 levels indicating efficacy in this species.

Lot number listed in the summary is the lot number of the vehicle, not the B2036-PEG.

Key finding(s):
1. There were no injection site reactions to the test agent in this rabbit study.
2. Efficacy was demonstrated by the determination that B2036-PEG clearly decreased serum IGF-1, indicating that the rabbit may be an appropriate species for further testing.

OVERALL SUMMARY AND EVALUATION:

Introduction:
Pre-IND meeting held with sponsor January 24, 1996. Alex Jordan and Ron Steigerwalt were pharmacologists present. Key toxicology items discussed: Dr. Jordan indicated that the monkey studies would probably be sufficient and also noted that carcinogenicity studies would eventually be required. Teratology should be performed in a species in which the drug works. There was a meeting with the sponsor regarding preclinical requirements for both acromegaly

indications. The CDER reproductive toxicology committee was consulted as to the appropriate design of studies. Joe DeGeorge was consulted regarding carcinogenicity studies. For Acromegaly, the Reproductive Toxicology committee recommended a modified Segment I/Segment II approach. The results of these consults were incorporated into a report of the minutes of the May 22, 1998 meeting. These comments were communicated to the sponsor in a fax on September 17, 1998.

The present submission provides a final report for the 28 day monkey toxicology study with every other day dosing. The only additional information provided in this report compared to the report provided in the initial submission were very limited toxicokinetic data, which are of little value. These are single point estimates and since the assay cross-reacts with monkey GH, it is not clear how reliable these estimations are. As with Growth Hormone products, the effect on IGF-1 might be a better way of assessing the effects of this product. Of note, however, are extensive missing pages in the overall report in the data sections. There does not appear to be a clear pattern as to why this occurred. In a previous amendment (SN034), studies were copied on both sides of the page. Many of the missing pages in this report could be due to the fact that this was intended to be copied in the same manner. However, there are some missing tables that would not fit in with such a pattern. Since this was previously reviewed, and this study was done with alternate day dosing, this study is not pivotal to the understanding of the toxicology of this compound. However, the sponsor should be alerted to be more careful in the preparation of subsequent amendments and the NDA. Another obvious error was the listing of the lot number of the vehicle in the summary of the rabbit study when the lot number of the study drug was being referenced.

A report on plasma levels from a number of mouse toxicology studies was also provided. It was hoped that this might clarify some confusion generated by the summary of study SEN-107 reviewed under serial number 034. However, although this is an extensive report on validation of the assay, the few actual data points that are listed are reported in a way so that the information cannot be linked to specific drug groups in any of the experiments. This report is of no use to the understanding of this compound.

The rabbit dermal study indicates that there was a definite response of IGF-1 to B2036-PEG. This suggests that the rabbit, unlike the mouse, might be a useful animal model. This is important to know since the sponsor has proposed doing pivotal NDA studies in the rabbit. The compound did not produce any injection site reactions in the rabbit.

Safety Evaluation: The main study submitted in this amendment was previously reviewed under the initial IND submission. The toxicokinetic data supplied in this "final" version do not provide any significant new information. The safety evaluation of this product has not changed from previous reviews.

Conclusions: The studies submitted in this amendment were incomplete. The 28 day monkey study was previously submitted and reviewed. This version is missing many pages. There were no significant additional findings in this report to add to the initial review.

RECOMMENDATIONS:

Internal comments: The requirements for toxicology studies to support NDA's for both the acromegaly [redacted] indications have been communicated to the sponsor (see copy of items above). No significant new preclinical data have been submitted since this communication.

External Recommendations (to sponsor): It should be pointed out to the sponsor that there were many pages missing in the report of the 28 day monkey study. Greater care needs to be taken in the preparation of future submissions.

Future development or NDA issues: Daily dosing for 3 months as described in previous reviews should help determine if there are any significant toxicities with this product. Of potential concern are hematological and bone effects as well as elevations in blood urea.

DRAFT LETTER CONTENT FOR SPONSOR:

There were an extensive number of pages and tables missing from the toxicology data section in the report of SEN-108 submitted under serial number 037 on March 8, 1999. There was no clear pattern to suggest why these pages might be missing. In addition, there were a number of small errors in summaries provided (for example, in the summary of study SEN-115, the lot number mentioned for B2036-PEG is actually the lot number of the vehicle). Greater care should be taken in future submissions to ensure that all data are provided and are correct. Submission pages should be numbered consecutively.

Reviewer signature/team leader signature [Concurrence/Non-concurrence]

Ronald W. Steigerwalt, Ph.D.
Pharmacology Team Leader

cc: IND Arch
HFD510
HFD510/Steigerwalt/CKing
Review Code: DE
Filename:
Appendix/attachments: Histopathology table

APPEARS THIS WAY
ON ORIGINAL

APPEARS THIS WAY
ON ORIGINAL

Addendum 1:Histopathology Inventory for IND [redacted]

STUDY	SEN-106 14 DAY IV	SEN-107 14 DAY SC	SEN-108 28 DAY QOD	SEN-109 26 WEEK SC WEEKLY
SPECIES	MOUSE	MOUSE	MONKEY	MONKEY
Adrenals	X*	X*	X*	X*
Aorta	X	X	X	
Bone Marrow smear				X
Bone (femur)	X	X	X	
Brain	X*	X*	X*	X*
Cecum	X	X		X
Cervix				
Colon	X	X		X
Duodenum	X	X		X
Epididymis	X	X		X
Esophagus	X	X		X
Eye	X	X		X
Fallopian tube				
Gall bladder	X	X		X
Gross lesions	X	X		X
Harderian gland				
Heart	X*	X*	X*	X*
Hyphophysis				
Ileum	X	X		X
Injection site	X	X		X
Jejunum	X	X		X
Kidneys	X*	X*	X*	X*
Lachrymal gland	X	X		
Larynx				
Liver	X*	X*	X*	X*
Lungs	X	X	X	X
Lymph nodes, cervical				
Lymph nodes mandibular				X
Lymph nodes, mesenteric	X	X		X
Mammary Gland	X	X		X
Nasal cavity				
Optic nerves	X	X		X
Ovaries	X*	X*	X*	X*
Pancreas	X	X		X
Parathyroid	X	X		X
Peripheral nerve				
Pharynx				
Pituitary	X	X	X*	X*
Prostate	X	X		X
Rectum	X	X		
Salivary gland	X	X		X
Sciatic nerve	X	X		
Seminal vesicles	X	X		X
Skeletal muscle	X	X		X
Skin	X	X		X
Spinal cord	X	X		X
Spleen	X*	X*	X*	X*
Sternum	X	X		X
Stomach	X	X		X
Testes	X*	X*	X*	X*
Thymus	X	X		X
Thyroid	X	X	X*	X*
Tongue	X	X		X
Trachea	X	X	X	X
Urinary bladder	X	X	X	X
Uterus	X	X	X	X
Vagina	X	X	X	
Zymbal gland				

* organ weight obtained

REVIEW AND EVALUATION OF PHARMACOLOGY/TOXICOLOGY DATA:

KEY WORDS: 26 Week Toxicology, Weekly dosing, Rhesus monkeys

Reviewer Name: Ronald W. Steigerwalt, Ph.D. Pharmacology Team Leader
Division Name: Division of Metabolic and Endocrine Drug Products (DMEDP)
HFD#510
Review Completion Date: March 1, 1999
Review number: 5

IND/NDA NUMBER: IND ——CROSS REFERENCE TO ——
Serial number/date/type of submission: SN#010, February 19, 1998 IT, CR
Information to sponsor: Yes () No (X)
Sponsor (or agent): Sensus Drug Development Corp.; Austin, TX 78701

DRUG
Code Name: B2036-PEG
Generic Name: rhGH antagonist (PEGylated)
Trade Name: Trovert™
Molecular Formula/ Molecular Weight: B2036-PEG is a 191 amino acid polypeptide with two disulfide bonds. As an analogue of hGH, it contains nine substitutions in the first and third α-helices. There are 4-5 PEG moieties (~5000 Da each) covalently bound to amino groups on the surface of the molecules. This increases its mass to 42-46,000Da.

Relevant INDs/NDAs/DMFs: IND ——

Drug Class: Modified recombinant human growth hormone.

Indication: Acromegaly (third line treatment)

Clinical formulation: Per vial:
10 mg B2036 protein
36 mg Mannitol
1.36 mg glycine
0.01M sodium phosphate, pH 7.4.
To be reconstituted in water up to 10 mg/ml (for clinical testing).

Route of administration: SC daily

Proposed clinical protocol or Use: No new protocols proposed in this submission. Initial studies attempted weekly administration of 80 mg. This has been increased to daily administration of 20 mg/day (total 140 mg/week). Individual unresponsive patients have received higher doses on a daily basis.

Previous clinical experience: Phase I: Placebo controlled, double blind, single rising dose study in healthy male volunteers was "nearing completion" at time of submission. B2036-PEG was "well-tolerated at doses of up to 1.0 mg/kg and efficacious in suppressing IGF-1 concentrations" at 0.3 mg/kg (3/6 pts) and 1.0 mg/kg (6/6 pts). No serious AE's; Other AE's resolved without sequelae. PK appeared to be linear within the 0.03 and 0.1 mg/kg groups. The PK were non-linear at effective doses of 0.3 and 1.0 mg/kg.

A single dose study in six acromegalic patients was initiated at [redacted] The [redacted] No data are available at this time.

Additional studies have assessed the daily administration of 20 mg/day in Acromegalic patients.

AE's: Preliminary data available:

0.6 mg/kg: (total 3 AE's)
1 subject unifocal ventricular extrasystoles before and after dosing. Extrasystoles remained 7 days later. Unlikely relationship with drug.

1 subject Abdominal pain and diarrhea (mild) Listed as remotely related to drug.
1 subject Headache (moderate). Listed as possibly related to drug.

0.1 mg/kg: 9 AE's: All mild: GI fullness, chest pressure, decreased appetite and headache considered remotely related to drug. Abdominal discomfort (possibly related) and nasal congestion and abnormal hunger were considered not related.

0.3 mg/kg: 3 AE's in 1 subject: rhinitis (day -3), oral herpes lesion (day -1). Mild itching at injection site lasting 12 h probably related to drug.

1.0mg/kg: 14 AE's: moderate epistaxis after minor trauma (remotely related) and headache (not related) 2 headaches were considered possibly related and parathesias of the tongue (2). Dry throat (2) and abdominal pain (1) were considered remotely related.

Phase 2: Protocol # SEN-3611: A Six Week, Double Blind, Placebo Controlled, Multiple Dose, Phase IIb study of B2036-PEG in the Treatment of Acromegaly. 45 patients (15/group) will receive weekly subcutaneous administrations for 6 weeks. Dose groups listed in protocol are 0, 30 and 80 mg of B2036-PEG. (Elsewhere in the submission, the dose selection is described as depending upon previous human experience). This was the initial plan, but the sponsor has since switched to daily dosing of 20 mg, with the possibility of increasing the dose further depending upon the patient response.

Disclaimer – use of sponsor's material: No sponsor material is reproduced in this report.

INTRODUCTION AND DRUG HISTORY:
Pre-IND meeting held with sponsor January 24, 1996. Alex Jordan and Ron Steigerwalt were pharmacologists present. Key toxicology items discussed: Dr. Jordan indicated that the monkey studies would probably be sufficient and also noted that carcinogenicity studies would eventually be required. Teratology should be performed in a species in which the drug works. There was a meeting with the sponsor regarding preclinical requirements for both acromegaly [redacted] indications. The CDER reproductive toxicology committee was consulted as to the appropriate design of studies. Joe DeGeorge was consulted regarding carcinogenicity studies. For Acromegaly, the Reproductive Toxicology committee recommended a modified Segment I/Segment II approach. The results of these consults were incorporated into a report of the minutes of the May 22, 1998 meeting (see copy of a summary of conclusions from the minutes from May 22, 1998 meeting with the sponsor. These comments were communicated to the sponsor in a fax on September 17, 1998.

The present submission provides the finalized report for the 26-week monkey toxicology study with weekly dosing. An interim (13-week) report was reviewed in the initial submission. This was

finalized on February 19, 1998, but was not previously reviewed since it deals with weekly dosing which is currently not relevant to the clinical situation.

Studies reviewed within this submission: SEN-109: B2036-PEG 26-Week Subcutaneous Toxicity Study in the Rhesus Monkey with an Interim Sacrifice after 13 Weeks Followed by an 8-Week Treatment-Free Period.
Studies not reviewed within this submission: None.

TOXICOLOGY:

General Comments: The interim report for 13-weeks was reviewed under the original IND submission. This constitutes a final report of the toxicology section of the 26-week study.

Study Title: B2036-PEG 26-Week Subcutaneous Toxicity Study in the Rhesus Monkey with an Interim Sacrifice after 13 Weeks Followed by an 8-Week Treatment-Free Period.
Study No: SEN-109
Amendment #10, Vol #1, and page #: 1
Conducting laboratory and location: [redacted]
Date of study initiation: October 30, 31, 1996.
GLP compliance: Yes (OECD)
QA- Report Yes (X) No ()

METHODS:
Dosing: SC WEEKLY

Dose (mg/kg)/week	Number of animals/time period					
	Week 13		Week 26		Week 34	
	males	females	males	Females	males	females
Control	2	2	4	4	2	2
0.3			4	4		
1			4	4		
3	2	2	4	4	2	2

species/strain: Captive bred Rhesus monkeys.
age: 2-3 years.
weight: 2.3-3.5 kg males, 2.2-3.7 kg females.
dosage groups in administered units: vehicle, 0.3, 1.0 and 3.0 mg/kg **dosed SC weekly**.
route, form, volume, and infusion rate: Dose volume 0.5 mL/kg/week, SC.
Drug, lot#, radiolabel, and % purity: 003007. Purity assumed 100% for dose calculations.
Formulation/vehicle: Nutropin buffer.

OBSERVATIONS AND TIMES:
Clinical signs: Morbidity/mortality checked twice daily, superficial clinical examinations daily. Full clinical exams during weeks 13, 26 and 34 prior to dosing. Injection sites examined on the day of dosing and the day after dosing.
Body weights: At least once weekly.
Food consumption: Daily.
Ophthalmoscopy: Pretest and weeks 13, 26 and 34.
Hematology: Pretest, then during weeks 12/13, 25/26, and 34. Hgb, MCH, MCHC, PCV, RBC, MCV, Retic, Plt, PT, APPT, tWBC, dif. WBC.
Clinical chemistry: Same as Hematology, Na, K, Cl, Ca, IP, Glu, Urea, Tchol, Tbili, Tprot, Alb, Glob, A/G ratio, Creat, ALP, AST, ALT, GGT
Urinalysis: Vol, Sp grav, appearance, pH, prot, glu, ketones, urobil., bili, blood, reducing substances, spun deposit, Ca

Gross pathology: terminal external surface, orifices, cranial cavity, external surface of brain and samples of spinal cord, thoracic and abdominal cavities and organs, cervical tissues and organs, carcass, injection sites.
Organs weighed: terminal see histopathology table
Histopathology: Terminal see histopathology table for listings
Toxicokinetics: Blood sampling on weeks 1, 13 and 26. h
Other: Sampling for hormone assays performed once pretest, then week 1, end of week 5, 13 , 26 and during week 34 (i.e., 5 days after dosing). Measured GH, IGF-1, Prolactin, Insulin, Antibodies (anti-GH and anti-test article). Note: animals were necropsied 6-7 days after the last dose for each time period.
RESULTS:
Clinical signs: No unscheduled deaths. Effects at injection were minor and infrequent, but occurred only in treated animals. These included:
Induration: 1 HD male (from week 6)
2 MD and 2 HD females (from 9th week)
Swelling: 1 MD male (from 10 weeks, bleeding noted at 20 weeks)
1 HD male from week 20
2 LD and 2 MD females (from 13/14 weeks)
1 MD female (from week 20)

One MD female vomited 20 min then 4h after dosing on day 1. Vomit found in cage on the morning of day 2. Since this didn't occur with other animals or higher doses, this was not considered treatment-related. Another finding of hair loss in one LD female was evident at week 13 and 24 (on arms at week 13, over most of body at week 25). One control male had hair loss on arms at week 24. One MD male had liquid diarrhea for 3 days during week 22. Other findings were present prior to dose administration and thus were not treatment-related.

Body weights: Body weight changes were variable. However, there was a definite trend for decrease in body weight (i.e., body weight LOSS compared to starting weight) in MD males and HD males and females. One MD female lost nearly as much body weight as the HD females. This was evident for these groups as early as the 13 week interim report. During the recovery period in males, body weights returned to control levels. While there was improvement in the females during the recovery period, body weights did not return to control levels. Since only 2 animals were examined in the recovery period/group, it is not clear if females would have fully recovered. Data suggest that they actually continued to lose weight compared to controls, but weighs did increase from the 26 week time point. There was no significant change in the low dose group for either sex.

Mean Body Weights Males (loss compared to control)				
Dose group (mg/kg)	DAY 1*	DAY 85*	Week 26 (n=4)	Recovery (n=2)
0	2.84	2.97	3.09	3.08
0.3	2.85	2.85 (-4%)	2.97 (-4%)	
1.0	3.02	2.70 (-9%)	2.58 (-17%)	
3.0	2.86	2.74 (-8%)	2.51 (-19%)	3.02 (-2%)
Mean Body Weights Females (loss compared to control)				
0	2.80	2.89	3.04	3.90
0.3	2.87	2.97	3.10	
1.0	2.79	2.82 (3%)	2.84 (-7%)	
3.0	2.74	2.54 (-13%)	2.26 (-26%)	2.55 (-35%)

*n=8; Day 85 data are presented since only 2 animals were presented at the 13 week time point.

Food consumption: Treated females (HD) tended to eat slightly less than controls from week 6. This reached statistical significance especially during weeks 11-13 and 25. There were some effects in the MD and LD females, but the differences were not statistically significant and probably not toxicologically significant. There were isolated incidences of decreased food consumption in HD males, but this was not consistent throughout the study and probably not due to test agent.

During recovery, food consumption was similar in all groups. It appears that the weight loss was not necessarily linked to changes in food consumption.

Ophthalmoscopy: No treatment-related findings. (Individual data were not presented.)

Hematology: HD animals of both sexes had **decreased Hb, PCV, and RBC** at weeks 13 and 26 (statistically significant in males at week 13 and females at week 26). Some evidence of a slight effect in MD females, but this is not clear). No change in circulating reticulocytes. Slight trend to lowered Platelets in MD and HD at both 13 and 26 weeks which was statistically significant for females in week 13. The significance of this is not clear since it was not significant at week 26 and non-significant effects were present pretest. Total WBC lower in treated females during weeks 13 and 26
HD had slightly shorter APTT at week 13 and 26 that were not clearly dose related since this was evident pretest.
Males recovered during the recovery period. One HD female still had low Hb, PCV and RBC (this might be related to the weight loss).

Mean Hematological Values (males) PRETEST					
Dose group (mg/kg)	Hb	RBC	PCV	APTT	TWBC
0	13.6	5.46	40.0	20.3	9.1
0.3	13.7	5.26	39.3	19.7	9.5
1.0	13.7	5.34	40.2	21.0	7.8
3.0	13.4	5.32	38.8	19.3	7.3
Mean Hematological Values (females) PRETEST					
0	13.1	5.30	38.4	21.3	9.5
0.3	13.7	5.39	39.5	19.8	7.2
1.0	12.9	5.33	38.5	22.0	7.8
3.0	13.3	5.35	39.3	21.4	8.0
Mean Hematological Values (males) WEEK 13					
0	13.8	5.72	40.6	20.6	8.9
0.3	13.5	5.44	39.9	20.1	9.9
1.0	13.5	5.51	39.4	19.9	9.0
3.0	12.6	**5.22***	**37.1***	19.6	8.7
Mean Hematological Values (females) WEEK 13					
0	13.4	5.49	39.2	18.6	9.3
0.3	13.5	5.46	39.9	19.3	**6.2***
1.0	13.2	5.41	38.5	17.3	**5.7***
3.0	12.8	5.23	37.3	17.8	**6.1***
Mean Hematological Values (males) WEEK 26					
0	13.6	5.51	40.3	20.7	8.8
0.3	13.3	5.28	39.4	20.0	8.1
1.0	13.7	5.50	40.4	19.4	6.0
3.0	12.6	5.29	37.8	**17.8***	7.6
Mean Hematological Values (females) WEEK 26					
0	13.7	5.70	41.3	21.2	10.1
0.3	13.0	5.34	38.8	20.4	**6.0***
1.0	12.6	5.26	37.6	18.8	**6.5***
3.0	12.3*	5.10	36.9	18.8	8.1

Clinical chemistry: MD males and HD males and females had **lower serum phosphorus** compared to concurrent controls and pretest values. These differences were less marked at 26 weeks compared to 13 weeks. This reached statistical significance in HD females at both week 13 and 26. (All female groups were lower than controls, but these were not statistically significant except at the HD). These were still slightly low at the end of the recovery period for HD animals. **Lower serum ALP** was also noted in treated animals. This was statistically significant at HD females and males and in LD and MD females in week 13. At week 26, this difference was only evident for HD females. **Blood urea was increased in HD males and MD and HD females.**

There was no notable difference after the recovery period. Cholesterol tended to be elevated in MD and HD groups for males and females at week 13 and 26 and was statistically significant for females at week 13.

Mean Clinical Chemistry Values (males) PRETEST			
Dose group (mg/kg)	P (mg/ml)	ALP (IU/L	Urea (g/L)
0	82	1608	.48
0.3	97	1313	.46
1.0	96	1287	.49
3.0	83	1205	.49
Mean Clinical Chemistry Values (females) PRETEST			
0	80	1674	.41
0.3	68	1383	.51
1.0	77	1114	.47
3.0	66	1250	.43
Mean Clinical Chemistry Values (males) WEEK 13			
0	81	2036	.53
0.3	75	1594	.57
1.0	66	1547	.58
3.0	61	1049	.65
Mean Clinical Chemistry Values (females) WEEK 13			
0	68	2388	.48
0.3	57	1731	.45
1.0	57	1307	.65
3.0	48	937	.61
Mean Clinical Chemistry Values (males) WEEK 26			
0	75	1547	.45
0.3	75	1211	.53
1.0	63	1047	.54
3.0	71	931	.63
Mean Clinical Chemistry Values (females) WEEK 26			
0	80	2058	.42
0.3	80	1655	.45
1.0	74	1516	.56
3.0	58	974	.61

Urinalysis: No treatment related changes.

Organ Weights: At 13 weeks, no clear treatment-related effects. However, in general, the absolute weights of livers of treated males and females tended to be lower than controls. Relative weights were also lower.
At 26 weeks, mean body weights of MD and HD males were statistically reduced compared to controls. This was also noted in HD females, but not statistically significant.
Adrenals: significant decrease in absolute and relative to brain weight in HD females. (significant decrease relative to body weight in MD and HD females.)
Kidney: Significant decrease in absolute and relative to brain weight in HD males.
Liver: absolute weights decreased non-significantly in HD males and females.
Pituitary relative to body weight significantly increased in MD, but not HD males
There was an increase in brain: body weight ratios in MD and HD animals which suggests that changes in liver or heart were a reflection of terminal bodyweights.

Absolute testes weights significantly reduced in MD and ovary weight were increased in LD but not when calculated as ratio of body weight. Given the fact that there was no dose dependence, these are not likely to be treatment-related.

Gross pathology: At 13 weeks, No clear treatment-related effects at either 13 or 26 weeks. However, at week 13, one HD female had adhesions and meningeal thickening. In one HD male, there was thickening at injection site, another HD male had small and pale spleen with dysmorphia. One HD male and female had raised areas in the jejunum. The female also had cystic area and adhesions in the liver, small ovaries, enlarged lymph nodes and dysmorphia of the spleen. There was a raised area in the spleen of one control female and one control male had fluid in the cranial cavity (this male also had dark/uncollapsed lungs and dark brain). These are mentioned primarily because they were noted in the HD group. However, at 26 weeks, there were only sporadic findings that occurred in controls and all dose groups, which were not treatment related. No treatment-related findings at recovery.

Histopathology: Note: no histopath data were presented in the 13 week interim report. Only 2 controls and 3 HD animals were evaluated at 13 weeks.
13 weeks: **Adrenals** of 3 HD males and females showed vacuolation of zona fasciculata. This was seen in only 1 control female. **Pancreas** in all 6 HD monkeys had "less prominent" Islets of Langerhans.
There was an **increase in fatty tissue** (fatty infiltration) in some organs, particularly at the injection site and intra-lobular deposits in the thymus and submucosal deposits in the colon and/or cecum. In males, **glycogen vacuolation in the liver** was present in controls and treated animals, but increased in level of severity in treated males. In females, this was present only in HD females.
A slight **reduction in bone of the body of the sternum and slight reduction in trabecular bone in the femur** in 2/3 HD males and females. There was also a diminution in the marrow of the femur, particularly in the head of the femur.
No obvious drug-related reaction at the injection site.
At 26 weeks (4/group analyzed). Again, **effects were noted in adrenals, bone and bone marrow** in treated animals. Increased fat in thymus, colon, cecum and injection site were less marked than at 13 weeks. The effects on the islet cells and glycogen content of the liver were present, but to a lesser extent (not obviously dose related) than at 13 weeks. The effect on adrenals was more apparently treatment-related in males (2 controls minimal, 3 LD males minimal to slight, 4 MD males slight, 3 HD males moderate). In females effects were similar in all groups, including controls. Pancreatic findings (less prominent islets) noted in one HD male, while another HD male had less staining of acinar cells. Thymic involution was noted in 2 males, 1 female of MD group and 2 HD males. Effect on trabecular bone in femur was minimal to slight in all MD males and all HD males and females. Reduction in bone in the body of the sternum was evident in these animals, but only in 3/4 HD females. Diminution of the marrow of the femur, particularly in the head was noted in MD and HD males and females.
After recovery period, effects on bone marrow were still noted, but other effects were not clearly evident. It should be noted that only 2 recovery animals in HD and controls were examined, therefore, the conclusions that can be drawn regarding recovery are not definitive.

Hormone measurements: IGF-1 concentrations were lower than controls in the MD and HD groups at weeks 1, 13 and 26. The differences were generally dose related and statistically significant (except for MD females on week 26. There were no findings at the LD group. Effect varied somewhat between groups and appeared to be more consistent throughout the study in males. The effect seemed to increase after week 1. In females, the effect was also strongest at week 1 in the MD group, the effect diminished with increasing duration in females. The effect was relatively consistent throughout the study for HD females.

IGF-1 concentrations, compared to controls (%):

GROUP/SEX	TIME POINT		
	WEEK 1	WEEK 13	WEEK 26
MD MALE	55	72	75
MD FEMALE	61	44	15
HD MALE	66	76	73
HD FEMALE	67	81	84

There was no effect on prolactin levels.
Insulin levels tended to be reduced, but since they were so variable, it is difficult to ascribe a specific treatment effect.

KEY STUDY FINDINGS:

1. Overall **NOAEL was 0.3 mg/kg on a weekly basis** (~ 3.6 mg/m^2/week).
2. There were no deaths. No injection site lesions were related to drug. No significant drug related clinical findings were noted.
3. There was a very definite, treatment-related decrease in **body weight loss** compared to starting weights in both MD and HD animals of both sexes. This appeared to recover in males after 8 weeks drug-free. The recovery was not clear in females. Although HD animals tended to eat less than controls, the change in food consumption was not apparently the full cause of weight loss.
4. At the HD, both sexes had **decreases in hemoglobin, packed cell volume and RBC counts.** There was also a trend for **decreased platelets** in the MD and HD. Males recovered from these changes, but one HD female still had low hemoglobin, PCV and RBC at the end of the 8 week recovery period. It is not clear if this was related to the weight changes.
5. MD males and HD males and females had **lower serum phosphorus and lower serum ALP. Blood urea was increased** in HD males and MD and HD females. These returned to baseline by the end of the recovery period. The findings for the urea were relatively small, but consistent throughout the study.
6. Histopathological changes were noted as follows:
   a. vacuolation of zona fasciculata of adrenals in HD males and females.
   b. "Less prominent" islets of Langerhans at HD all animals.
   c. Increase in fatty infiltration in colon, cecum, thymus
   d. An increase in glycogen vacuolation in the liver
   e. Reduction in bone of the body of the sternum and slight reduction in trabecular bone in the femur. This was accompanied by diminution of the marrow of the femur, particularly in the head of MD and HD animals.
   f. Bone effects were still noted in the recovery animals.
7. IGF-1 was clearly lower than controls in MD and HD groups at 1, 13 and 26 weeks. There was no effect on prolactin levels. Insulin levels tended to be reduced, but since they were so variable, it is difficult to ascribe a specific treatment effect.

OVERALL SUMMARY AND EVALUATION:

Introduction:
Pre-IND meeting held with sponsor January 24, 1996. Alex Jordan and Ron Steigerwalt were pharmacologists present. Key toxicology items discussed: Dr. Jordan indicated that the monkey studies would probably be sufficient and also noted that carcinogenicity studies would eventually be required. Teratology should be performed in a species in which the drug works. There was a meeting with the sponsor regarding preclinical requirements for both acromegaly indications. The CDER reproductive toxicology committee was consulted as to the appropriate design of studies. Joe DeGeorge was consulted regarding carcinogenicity studies. For Acromegaly, the Reproductive Toxicology committee recommended a modified Segment I/Segment II approach. The results of these consults were incorporated into a report of the

minutes of the May 22, 1998 meeting (see copy of a summary of conclusions from the minutes from May 22, 1998 meeting with the sponsor. These comments were communicated to the sponsor in a fax on September 17, 1998.

The present submission provides a final report for the 26 week monkey toxicology study with weekly dosing.

Safety Evaluation: The sponsor has switched from the initially proposed weekly dosing to daily dosing. The exposure for daily dosing in humans on a weekly basis exceeds the single dose/week initially proposed by nearly 2-fold the highest exposure in the animal studies. The highest exposure in the animal studies approximates the proposed human exposure. Toxicity findings appeared to be relatively minor and there were no kidney findings in the multiple daily dose studies in mice or monkeys (see original review) which would suggest that there is not a problem with kidney effects noted in animal studies with a PEGylated TNF. The sponsor has been allowed to proceed in human trials without specific coverage of daily dosing in animal studies of equal duration. This was communicated to the sponsor in a fax. The specific points are reproduced after the conclusion section of this document. Particular concerns relate to changes in hematological parameters and bone effects. Given the effects on adrenals, it might be useful to evaluate changes in adrenal function in normal subjects particularly for the ——— indication.

Clinical Relevance of Safety Issues: No significant safety issues are evident. There were some effects that might be related to the GH antagonism (e.g., bone effects, possibly some hematological effects). This may not be a significant concern in acromegaly patients since the point to therapy is to block excessive GH effect rather than decrease the effect below physiological levels. ——— The sponsor is dosing humans daily beyond duration covered by animal toxicity studies. Highest exposures in animal toxicology studies approximate the human exposure when calculated on a mg/m^2 basis. It is not clear what actual plasma level multiples would be. Blood urea levels rose slightly in treated animals and BUN should be watched carefully in clinical trials.

Conclusions: The study presented is a final report of a toxicology study based upon weekly dosing in monkeys. This has limited relevance to the current clinical dosing. This submission does not add significantly to the overall understanding of the toxicity of this product based on daily dosing, but does indicate that with chronic dosing, bone and hematological parameters should be monitored. The sponsor basically dismisses some hematological findings that nonetheless were consistent between mouse and monkey studies. These were not severe toxic findings, and can be monitored in clinical trials. If this is due to the pharmacodynamic effects of GH antagonism, there may be little concern for acromegaly patients for whom the goal is to reduce GH activity to normal levels. ———

STUDIES REQUESTED FOR B2036-PEG (FROM MEETING MINUTES OF MAY 22 1998 MEETING-MINUTES FINALIZED AUGUST 10,1998 AFTER COMMITTEE CONSULTATIONS) THESE HAVE BEEN COMMUNICATED TO THE SPONSOR

The following provides an outline for the preclinical studies necessary for filing of an NDA for Trovert:

1. Toxicity studies: A three-month toxicity study in rats with daily dosing and a minimum high dose of 10-fold (preferably 25-fold) the human maximum AUC will be provided for the filing of an acromegaly indication. ———

sensitivity to tissue accumulation. These studies should be performed with the clinical formulation, if possible.

2. Reproductive toxicity: For the acromegaly indication, the use of the rabbit as a single species in the reproductive toxicity studies should suffice. However, the Reproductive Toxicology Committee recommended that a modified Seg I/Seg II approach be undertaken, in which one set of experiments begins treatment of inseminated does on the day of insemination, and continues treatment through Day 7; and the second set of experiments treats during the traditional organogenesis period (Days 6-19).

The Division concurs with the committee recommendations.

3. Carcinogenicity studies: The sponsor should provide the following at the NDA submission for acromegaly: GH binding studies as outlined at the May 22 meeting, and the PC-3 tumor cell transplant study proposed by the sponsor during the 6/8/98 teleconference. A single carcinogenicity study submitted Phase IV could cover the acromegaly indication.

It should be understood that the above recommendations are based on the proposal for daily clinical dosing with Trovert™ at 20 mg/kg/day. Changes in the clinical proposal or formulation could necessitate additional preclinical studies.

RECOMMENDATIONS:

Internal comments: The requirements for toxicology studies to support NDA's for both the acromegaly indications have been communicated to the sponsor (see copy of items above). No additional preclinical data have been submitted since this communication.

External Recommendations (to sponsor): no action indicated at this time.

Future development or NDA issues: Daily dosing for 3 months as described in the attached recommendations should help determine if there are any significant toxicities with this product. Of potential concern are hematological and bone effects as well as elevations in blood urea.

Reviewer signature/team leader signature [Concurrence/Non-concurrence]

Ronald W. Steigerwalt, Ph.D.
Pharmacology Team Leader

cc: IND Arch
HFD510
HFD510/Steigerwalt/CKing/Perlstein
Review Code: ND
Filename:

Appendix/attachments:
3. Histopathology table

Addendum 1: Histopathology Inventory for IND

STUDY	SEN-106 14 DAY IV	SEN-107 14 DAY SC	SEN-109 26 WEEK SC WEEKLY
SPECIES	MOUSE	MOUSE	MONKEY
Adrenals	X*	X*	X*
Aorta	X	X	
Bone Marrow smear			X
Bone (femur)	X	X	
Brain	X*	X*	X*
Cecum	X	X	X
Cervix			
Colon	X	X	X
Duodenum	X	X	X
Epididymis	X	X	X
Esophagus	X	X	X
Eye	X	X	X
Fallopian tube			
Gall bladder	X	X	X
Gross lesions	X	X	X
Harderian gland			
Heart	X*	X*	X*
Hyphophysis			
Ileum	X	X	X
Injection site	X	X	X
Jejunum	X	X	X
Kidneys	X*	X*	X*
Lachrymal gland	X	X	
Larynx			
Liver	X*	X*	X*
Lungs	X	X	X
Lymph nodes, cervical			
Lymph nodes mandibular			X
Lymph nodes, mesenteric	X	X	X
Mammary Gland	X	X	X
Nasal cavity			
Optic nerves	X	X	X
Ovaries	X*	X*	X*
Pancreas	X	X	X
Parathyroid	X	X	X
Peripheral nerve			
Pharynx			
Pituitary	X	X	X*
Prostate	X	X	X
Rectum	X	X	
Salivary gland	X	X	X
Sciatic nerve	X	X	
Seminal vesicles	X	X	X
Skeletal muscle	X	X	X
Skin	X	X	X
Spinal cord	X	X	X
Spleen	X*	X*	X*
Sternum	X	X	X
Stomach	X	X	X
Testes	X*	X*	X*
Thymus	X	X	X
Thyroid	X	X	X*
Tongue	X	X	X
Trachea	X	X	X
Urinary bladder	X	X	X
Uterus	X	X	X
Vagina	X	X	
Zymbal gland			

* organ weight obtained

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature.

/s/

Fred Alavi
4/27/01 01:18:07 PM
PHARMACOLOGIST
Pharm/Tox Review, NDA 21-106, pegvisomant, a growth hormone antagonist for treatment of acromegaly
Jeri, I made the corrections. I think it is ready for sign off.

Jeri El Hage
4/30/01 10:48:58 AM
PHARMACOLOGIST
Crystal - Please see list of recommendations to sponsor.